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Research Article

Phytochemical Profile and Antioxidant Potential of *Berberis aristata* Bark in Diabetes Mellitus Management

Vivek Dwivedi [1,2*], Shahnawaz Sameem [3], Amita Verma [1]

[1] *Bioorganic and Medicinal Chemistry Research Laboratory, Department of Pharmaceutical Sciences, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, 211007, India*

[2] *Chandra Shekhar Singh College of Pharmacy, Koilaha, Kaushambi, 212213, India*

[3] *Medicinal Chemistry Research Laboratory, Faculty of Pharmacy, United University, Prayagraj, 211012, India*

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*Address for Correspondence:

Vivek Dwivedi, Bioorganic and Medicinal Chemistry Research Laboratory, Department of Pharmaceutical Sciences, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj, 211007, India

Abstract

Diabetes mellitus is a long known metabolic disorder that is afflicting millions of people all over the world. High levels of blood sugar (also known as glucose) in the blood that can develop into other health problems if untreated, this is the main feature. In this case the body cannot make enough insulin to regulate blood sugar or does not use insulin properly. Diabetes have two types, i.e. diabetes 1st type and diabetes 2nd type. Chronic diseases both are called diabetes that affect how glucose is processed in the body causing blood sugar levels to be high. They however have different causes, symptoms, and treatment options.A plant of the Berberidaceae family, Berberis aristata is often called Indian Barberry or Tree Turmeric. The traditional use of Berberis aristata has been very long. One of its many active ingredients, berberine has shown antimicrobial, anti inflammatory as well as antioxidant and antidiabetic properties. The plant is used for treating numerous medical conditions, e.g. digestive problems, respiratory infections, skin conditions, diabetes. The Berberis aristata in ayurvedic medicine is used in diabetes and diabetes related complications. It is understood to improve insulin sensitivity and to promote glucose uptake by the cells to help with blood sugar levels. Berberis aristata is a long traditional medicine. Antimicrobial, antidiabetic, antioxidant and anti-inflammatory properties have been demonstrated for one of its many active ingredients, berberine, although it is not believed to be the most important one when it comes to diabetes. The plant is used for treatment of numerous medical conditions such as digestive, respiratory infection, skin, and diabetes.

Keywords: Diabetes mellitus, Antioxidant, Berberine, Berberis aristata

INTRODUCTION

Indian Barberry or Tree Turmeric (*Berberis aristata*), is an Ayurvedic, as well as Unani and Traditional Chinese Medicine (TCM), medicinal plant of Berberidaceae family with therapeutic properties [13, 14, 25]. This plant is rich in bioactive alkaloids, especially berberine, and has been cited to possess potential in cures of metabolic disorders, especially Diabetes Mellitus (DM). Previous modern pharmacological studies have validated its antidiabetic, anti-inflammatory, antioxidant and lipid lowering effects which are traditionally indicated for use in gastrointestinal ailments, microbial infections and inflammatory conditions [1, 6, 14]. The main bioactive compound in berberine is berberine, from which it is thought to exert its glucose metabolism enhancing, insulin sensitizing, AMPK activating, and insulin resisting as well as Hepatoprotective effect and cardioprotective effects by lowering cholesterol and triglycerides [8, 15]. Furthermore, its anti-inflammatory and an antioxidant mechanisms relieve oxidative stress and chronic inflammation, which are important in the progression and complications of diabetes like nephropathy, neuropathy, and retinopathy [19,20,22,25]. In addition, it may act by altering lipid metabolism and adipogenesis and thus facilitate weight management, an important T2DM prevention and control factor [2, 3]. Berberis aristata has been a target of more recent clinical investigations and appears to have the potential to regulate glycemia as well as postprandial glucose control and diabetes-related complications, thus making it a promising phytotherapeutic agent [4, 11, 24]. In view of a growing diabetic and metabolic disorder burdens, which are currently burdening world population this review focuses on its bioactive compounds, pharmacological mechanisms, and clinical applications toward understanding its therapeuticals role and future

research directions in integrating them into modern pharmacotherapies [5, 26].

2. MATERIAL AND METHODS

2.1Plant resources and preparation: Barberis aristata bark were harvested from Indian plant species, verified, and dried at room temperature (25–30ºC) for several days. The 200g of bark were then meticulously separated from the stem and seeds by hand-picking, and finally ground using an electronic grinder. The powdered extract was kept in sealed polyethylene bags for storage [17, 18].

2.2Extraction of Phytochemical / Phytoconstituent of Barberis aristata

The soxhlet apparatus extraction technique is used to extract the phytoconstituents found in barberis aristata bark (Fig.1). First, the crude powder of the leaves is packed into a thimble and attached to a round-bottom flask. Next, ethanol is added as a solvent from the top of the condenser, which is attached to the upper side of the thimble to a round-bottom flask and placed onto a heating mantle. Finally, the soxhlet extractor is attached above the flask, followed by a reflex condenser that has cold water entering at the bottom and exiting above the extractor. Using solvent ethanol, for extracting the phytochemicals/phytoconstituents [17].

Figure 1: Soxhlet Appratus

Barberis aristata

↓

desired bark and powdered to desire size

↓

Powdered drug was filled in Soxhlet apparatus for Pet. Ether extraction

↓

Defatted extract was collected and dried Marc was shade dried and

↓

refilled in soxhlet for ethanolic extraction Extract was collected

↓

and dried by evaporating the solvent in dessicator All extract were stored for further study

2.3 Phytochemical Analysis

The aim of the phytochemical analysis was to identify the alkaloids, glycosides, flavonoids, volatile oils, balsams, terpenes, tannins, phenols, and resins that were found in the crude leaf extract of barberis aristata [10].

2.3.1 Test for Alkaloids (Mayer's Test) (Khandelwal,2017)

Alkaloids were identified by mixing two ml of ethanolic crude extract of cannabis sativa with a few drops of Mayer's reagent gives precipitate [27].

Test For Alkaloids (Dragendorff's Test) (Khandelwal,2017)

Alkaloids were identified by mixing two ml of ethanolic crude extract of barberis aristatawith a few drops of dragendorff's reagent gives orange brown precipitate [27].

2.3.2 Test ForSaponins (Foam test) (Khandelwal, 2017)

To determine saponins, shake the ethanolic extract of with water.Persistent stable foam observed [27].

2.3.3 Test for Tannins (FeCl3 test) (Khandelwal, 2017)

To measure tannins, mix 2 ml of ethanolic crude leaf extract of barberis aristata in a test tube it gives deep blue-black colour [27].

2.3.4Test For Flavonoids (Sulphuric acid test) (Khandelwal, 2010)

To detect flavonoids, 2 ml of ethanolic crude extract of and add sulphuric acid it gives deep yellow solution [27].

2.3.5 Test for Cardiac glycosides (kellerkilliani test) (Khandelwal, 2017)

To this mix, 1 drop of 5% ferric chloride and conc. H2SO4 is added and 2 ml of glacial acetic acid as well as 2 ml ethanolic crude bark extract is mixed in a test tube. On mixing two liquids layer, reddish brown appears at the junction and upper layer is bluish green [27].

2.3.6 Test ForTerpenes(Salkowski test) (Khandelwal, 2017)

To detect terpenes, 5ml of ethanolic extract is mixed with 2 ml of chloroform and 3 ml of conc. $H_2SO_4$is added to form a layer. A reddish brown colouration of the interface is formed [27].

2.3.7 Test for Volatile Oil (Khandelwal, 2017)

Solubility test: Volatile oils are soluble in 90% alchohol [27].

2.3.8 Test For Resin –

To detect the presence of resin, add 2 ml of acetic anhydride with 2 ml of ethanolic bark extract in an test tube. Add 3 drops of concentratedsulphuric acidand it gives a violet colour shift [27].

3. RESULT AND DISCUSSION

3.1 Phytochemical analysis

Table: 1- Phytochemical Screening of the Ethanolic Extract of Barberis aristata

S.No.	Phytochemical Constituent	Ethanolic Extract
1	Test for glycosides Keller Killani test	Positive
2	Test for Alkaloid Mayers's test Dragondroff's test	Positive
3	Test for volatile oils	Positive
4	Test for Steroids	Positive
5	Test for flavanoids Alkaline test	Positive
6	Test for Carbohydrates Molisch's test Benedict test Fehling test	 Positive Positive Positive
7	Test for Starch	Negative
8	Test for Saponins	Positive

3.2 Characterization of Barberis aristata (Thin Layer Chromatography)

Thin layer chromatography (TLC) is mainly based at the working of adsorption. This type of chromatography uses the cellulosic segment that consists of the dissolved solutes transporting through the floor of the stationary section. Skinny layer chromatography (TLC) of each solvent extract was brought about by the use of silica gel 60F254, 7X6 cm (Merck), cut the use of not unusual domestic scissors, in step with the vintage one-dimensional ascending process. It was used to make plate marks with soft pencil, and was soft itself. Glass capillaries were utilised to identify the sample for TLC.The TLC chamber was developed using a mixture of Toluene: Ethyl acetate (8:2) For improvement, pre-saturation with mobile segment for 20 mins turned into employed. To discover the bands on the TLC plates, newly produced iodine reagents were hired after the run plates had been dried and sprayed. The retention aspect (Rf) became used to explicit the energetic compound's mobility, and values were decided for numerous samples [9, 10, 16].

Detection and Calculation of Rf. Value

Once the chromatogram was developed the Rf Value (Fig.2) (Table. 2) of the spot was calculated using the formula an

R_f = Distance travelled by solute / Distance travelled by solvent

Table 2: Rf value of Mobile Phase

S.No.	Mobile Phase	Rf value
1.	Toluene:Ethylacetate (8:2) Dis.travelbymobile phase=4.5cm	=0.11,0.2,0.95

Figure 2: Toluene:Ethylacetate(8:2)

3.3 In-vitro antioxidant potential of Barberis aristata

Different stock of the test compound (according to the mention in excel sheet) was added to 0.1 ml of 0.1mM DPPH solution (SRL Chem – Cat no.– SR-29128) in Methanol (SD fine- Cat no.- 10930lC250) in a 96 well plate 10µl of the test compound. standard – Ascorbic Acid (SRL, Cat no- 23006) was used in the rection quadruplicate form with duplicate blank containing 0.2 ml of Methanol and 10µl standard/sample at varying concentrations (According to excel sheet) of Ascorbic Acid as per table mention. The wells without treatment were controlled and the wells without reagent was taken as Blank.

Light was prevented from falling on the plate for 30 min. As you can see, at the end of the incubation, the decolourisation was read at 517 nm using a micro plate reader (iMark, BioRad). Control was served by Reaction mixture of 20µl deionized water. '% inhibition' with respect to control was presented as its scavenging activity, and IC50 was calculated with the help of Software Graph Pad Prism 6. A graph was made which was of X axis (Sample Concentration) Vs. Y axis (% inhibition wrt control) [7].

Calculations

$$\% \text{ RSA} = ((\text{Abs}_{\text{Control}} - \text{Abs}_{\text{Sample}})/\text{Abs}_{\text{Control}}) \times 100$$

RSA = Radical Scavenging Activity

$\text{Abs}_{\text{Control}}$ = Absorbance of control

$\text{Abs}_{\text{Sample}}$ = Absorbance of sample

Result

Table 2

Sample code	IC_{50}value(µg/ml) (Mean± SEM)
Ascorbic Acid	15.2 ± 0.024
Barberis aristata	236.7 ± 0.051

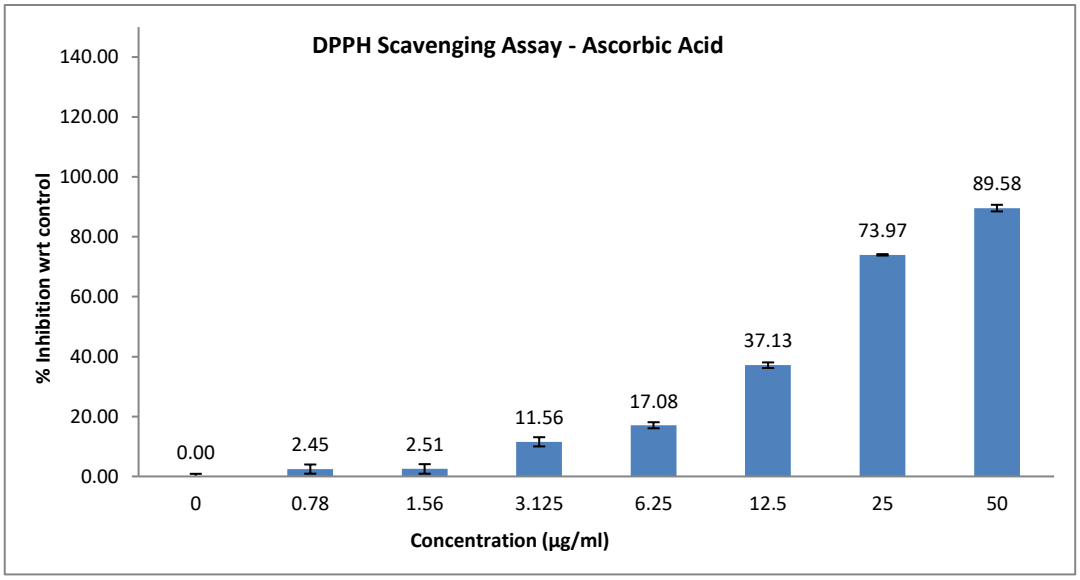

Figure 3: DPPH Scavenging assay of Ascorbic acid

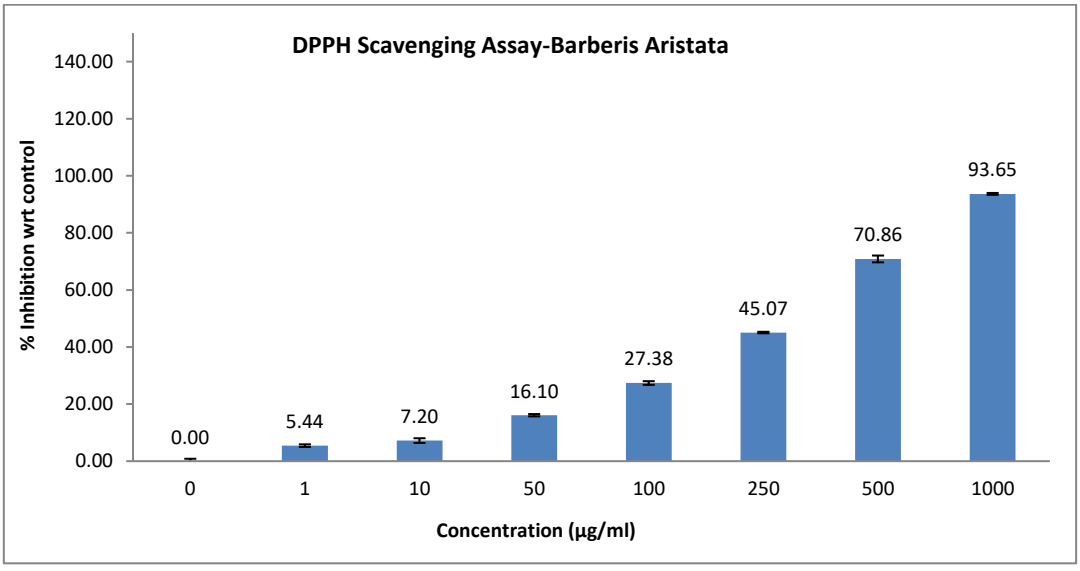

Figure 4: DPPH Scavenging assay of Barberis aristata

CONCLUSION

In summary, the phytochemical analysis and characterization using TLC and the extraction technique utilized ethanol extract for pharmacological investigation as an antioxidant in diabetic mellitus. In-vitro assays revealed significant scavenging activity by the barberis aristata bark ethanolic extract compared to the standard ascorbic acid.

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