Natural Therapeutics from *Cissus quadrangularis*: Phytochemical Constituents and Their Role in Bone Regeneration

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Abstract- Cissus quadrangularis, commonly known as Hadjod, is a traditional medicinal plant that has been used for many years, especially in Ayurveda, for healing broken bones and improving overall health. This review brings together information from different studies to better understand its chemical composition and health benefits.

Hadjod contains several important natural compounds like flavonoids, tannins, and vitamin C, which help the body fight damage caused by harmful molecules (free radicals). Because of these compounds, the plant shows strong antioxidants and anti-inflammatory effects. Research studies have also shown that Hadjod can help in faster healing of bone fractures, reduce pain, and support bone strength.

Apart from bone health, Hadjod may also be helpful in managing problems like obesity, stomach ulcers, and some metabolic disorders. Its effects are linked to how it protects body cells, reduces inflammation, and stabilizes proteins in the body.

Key words: Cissus quadrangularis, Hadjod, Phytochemicals, Antioxidant activity, Anti-inflammatory activity, Bone fracture healing, Medicinal plants

I. INTRODUCTION

Medicinal plants are the ultimate treasure trove of drugs for traditional medicine, modern pharmaceuticals, nutraceuticals, food supplements, and even the building blocks for synthetic drugs. India is blessed with a wealth of native herbal resources—around 20,000 plant species, about 2,500 of which pack serious medicinal punch. The magic lies in their phytochemicals, like flavonoids, polyphenols, alkaloids, tannins, glycosides, and saponins, stashed away in leaves, bark, stems, seeds, roots, fruits, and more. These compounds deliver powerful antioxidant, antimicrobial, and anti-inflammatory effects, giving them endless healing potential. They tackle tough conditions like cancer, diabetes, and arthritis without the nasty side effects of conventional drugs—truly "friendly medicines." Take Cissus quadrangularis, for example: it's a standout plant researcher are exploring for bone health issues, obesity, gastrointestinal problems, and beyond.

Hadjod (Cissus quadrangularis), the famous "Bonesetter," is a powerhouse Ayurvedic shrub that's a go-to for speeding up bone fracture healing, easing joint pain, and fighting osteoporosis. Packed with calcium, antioxidants, and steroidal compounds, it strengthens ligaments and can slash fracture recovery time by 40-50%. It promotes the natural union of fractured bones (Sandhaniya) and ramps up bone density.Great for osteoarthritis, rheumatoid arthritis, weak bones, and everyday pain relief. Helps with weight management, soothes gastric issues and ulcers, and tamps down inflammation. Usually as powder, tablets, or capsules. It’s generally safe, but overdoing it might lead to headaches, dry mouth, or diarrhea. Pair it with milk or ghee for better absorption. Always check with a doctor for the right dose, especially if using it long-term.

II. MATERIAL AND METHODS

Plant Extract Preparation: The stems (Figure 2) of Hadjod plant were peeled, chopped and grinded in ssuitable solvent with the help of mortar and pestle. The extracts of the stem were prepared by soaking 20 gm of finely ground stem in 80 ml of ethanolic and

methanolic solvents separately for 12 hours. The extracts were then filtered using Whatman filter paper No.42. Ccollected filtrates were used for carrying out different bbiochemical assays.

Phytochemical Test

Screening for phytochemicals in the plant reveals a wealth of compounds, including alkaloids, steroids, tannins, saponins, phytoestrogen steroids, cardio glycosides, and terpenoids. Flavonoids from CQ pack a punch with their antimicrobial, anti-inflammatory, anti-diarrheal, and anticancer effects [24–26]. Terpenoids shine as anti-tumor agents too—they target enzymes like lipoxygenase, phospholipases, and cyclooxygenase, and they're key players in fighting inflammation. Studies on stilbenes show their powerhouse anti-cancer and anti-inflammatory compounds that act as estrogen receptor α agonists and effectively trigger apoptosis. Plants produce primary metabolites to meet their basic needs, while secondary metabolites step in for self-defense. On top of that, this plant is loaded with minerals like calcium, potassium, iron, copper, cadmium, calcium oxalate, and zinc; heavy metals like lead; plus, phenols, vitamins, carotenes, and more.

Test for flavonoids

The alkaline reagent test is a straightforward qualitative method to spot flavonoids in plant extracts. It works because flavonoids—with their phenolic hydroxyl groups—react with alkaline solutions like sodium hydroxide (NaOH) to form yellow-colored phenolate ions

Total Flavonoid Content: We measured the total flavonoid content (TFC) in the stem extract of the Hadjod plant using the conventional aluminium chloride method. Initially, we combined 0.1 ml of the test sample with 0.1 ml of 1M potassium acetate solution, 0.1 ml of 10% aluminium chloride, and 1 ml of distilled water. To get a total volume of 3.3 ml, we then added 2 ml more distilled water. We measured absorbance at 415 nm after vigorously shaking the mixture and incubating it for 30 minutes. We used quercetin concentrations ranging from 100 to 500 µg/ml against a blank to create a standard plot at 415 nm for calibration. Lastly, we determined the amounts of flavonoids in the samples from this plot.

Test for phenols

A straightforward qualitative test for phenolic compounds, such as flavonoids, in plant extracts is the ferric chloride test. Depending on the particular phenolic compound present, it produces a characteristic green, blue, purple, or black color

Total Phenolic Content:.We used the Folin-Ciocalteu reagent to determine the total phenol content. We thoroughly mixed 1 ml of distilled water, 5 ml of 10% Folin-Ciocalteu reagent, and 4 ml of 1M sodium carbonate with 100 µl of extract. We used a spectrophotometer to measure the absorbance at 750 nm in comparison to a blank after 30 minutes of room temperature incubation. Gallic acid at 750 nm (concentrations ranging from 25 to 200 µg/ml) was used to create a calibration curve. This curve was used to calculate the phenolic concentrations in the samples, which were then expressed as µg of gallic acid equivalents (GAE) per gram of extract. Every measurement was carried out three times.

Test for terpenoids

A common qualitative technique for identifying terpenoids in plant extracts is the Salkowski test. It depends on the reaction of sulfuric acid concentrated terpenoids to produce colorful complexes and a distinctive color shift.

Test for glycosides:

We combined 5 ml of each extract with 2 ml of glacial acetic acid and 1 ml of 5% ferric chloride to test glycosides. We moved it to a test tube with two milliliters of concentrated H_2SO_4 after gently heating it. Glycosides are confirmed by a bluish-green color in the acetic acid layer and a reddish-brown color at the intersection of the two layers.

Test for tannins:

Braymer's test is a simple chemical test used to check if tannins are present in plant extracts. In this method, a small amount of the extract is mixed with a few drops of ferric chloride solution. If hydrolysable tannins are present, the solution turns blue-black, while condensed tannins give a greenish-black color.

Test for steroids:

A straightforward technique for identifying sterols, such as cholesterol, in a sample is the Salkowski test.

In this test, concentrated sulfuric acid is carefully added after the sample has been dissolved in chloroform. The sulfuric acid layer fluoresces yellow to green when sterols are present, while the chloroform layer turns red or reddish-brown. The sterol and acid react to form structures with conjugated double bonds, which causes this color shift.

Test for saponins:

A straightforward technique for detecting saponins, particularly in plant extracts, is the foam test. This test involves mixing a tiny quantity of the extract with water in a test tube and giving it a good shake. A stable foam or froth forms on the surface if saponins are present. This occurs because of saponins' soap-like qualities, which lower surface tension and produce bubbles. The foam test is a popular method for quickly and accurately identifying saponins in pharmacognosy and phytochemical screening.

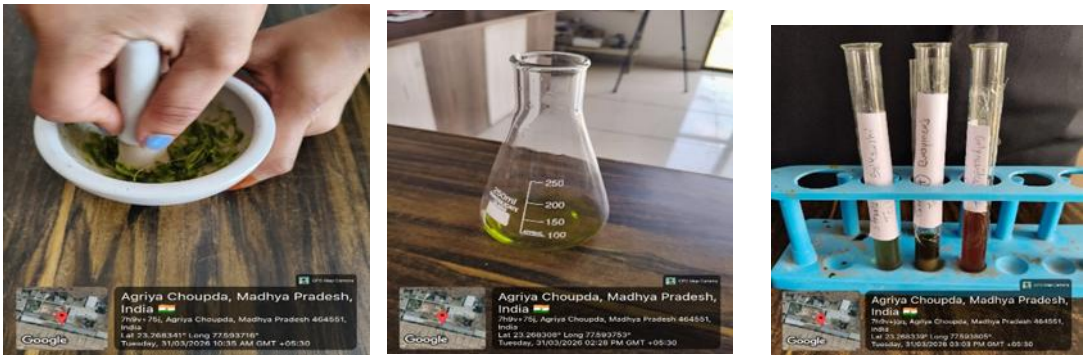

Figure: 1 Phytochemical Screening of *Cissus quadrangularis* Stem Extract

Table 1: Qualitative Phytochemical Screening of *Cissus quadrangularis* Stem Extract

S.No	Metabolite	Test	Experiment	Observation
1.	Flavonoids	Alkaline Reagent ATest	2-3 ml extract + 2 ml 40% NaOH	Deep yellow colour appears
2.	Terpenoids	Salkowsky's Test	2 ml extract + 1 ml chloroform + few drops of conc, H2SO4	Reddish-brown colouration appears at the interface
3.	Glycosides	Kellarkialliani	5 ml extract + 2 ml glacial acetic acid + 1 ml 5% FeCl3 + heat carefully then cool +transfer it to a TT containing 2 ml conc. H2SO4	Reddish- brown and greenish- blue rings appear at the junctions
4.	Saponins	Foam Test	2 ml extract + 5 ml D/W + shake TT	Stable foam
5.	Tannins	Braymer's Test	1ml extract + 2 ml of 10% FeCl3	Dark blue colour appears
6.	Phenols	Ferric chloride Test	1-2 ml extract + 1 ml of 5 % FeCl3	Deep blue colour appears
7.	Steroids	Salkowsky's Test	1 ml extract + 1 ml chloroform +1 ml conc. H2SO4 along the sides of test tube	Chloroform layer appears red and acid layer shows greenish yellow colour

Thin layer chromatography:

We performed thin-layer chromatography (TLC) using homemade silica gel G plates (400 mesh size) prepared in the lab. Samples were spotted 2 cm from the bottom of the plates with micropipettes for even application, then allowed to dry. The plates were developed in a chromatography chamber with a chloroform:methanol solvent system (15:1 ratio). After air-drying, we baked them in a hot air oven at 100°C for 5-6 minutes, then observed and visualized the spots under visible light. Retention factor (Rf)

values for each active compound were calculated based on visible light observations.

III. RESULTS AND DISCUSSION

Hadjod's phytochemical analysis revealed the presence of a number of significant bioactive substances. The following was verified by various tests:

Table 2: Qualitative phytochemical analysis of stem of Hadjod plant

S.No.	Cissus quadrangularis	Ethanolic Stem Extract
1	Triterpenoids and steroids	present
2	Alkaloids	present
3	Flavonoids	present
4	Tannins	present
5	Saponins	present
6	Glycosides	present
7	Phenolic substances	present

Depending on the type of extract (alcoholic or aqueous), certain compounds like proteins and carbohydrates may also be found.

The results clearly show that Hadjod is rich in natural chemical compounds that are beneficial for health. These compounds explain why this plant has been used in traditional medicine for a long time.

Flavonoids and phenolic compounds are known for their antioxidant activity, which helps protect the body from damage caused by free radicals. This supports the plant's role in healing and reducing inflammation.

Alkaloids often have pain-relieving and medicinal properties, which may help in reducing discomfort during bone injuries.

Tannins have astringent properties, which help in wound healing and prevent infections.

Saponins are known to support bone health and immune function, which is important because Hadjod is widely used for bone fracture healing.

Steroids and triterpenoids may contribute to anti-inflammatory effects and help in faster recovery of damaged tissues.

IV. DISCUSSION

Our in vitro studies on the ethanolic stem extract of the Hadjod plant show that it possesses strong antioxidant and anti-inflammatory properties, suggesting that it could serve as a promising source for developing anti-inflammatory Drugs. to better understand its anti-inflammatory effects, we performed several tests,

In which we indicate the presence of Tannis (Braymer test), Steroids (Salkowshi Test), and Glycosides. The findings from these tests indicate that the ethanolic stem extract of Hadjod has significant potential as an anti-inflammatory agent. Flavonoids and other polyphenols play a major role in the antioxidant and anti-inflammatory activities of many plants. Therefore, the observed effects of the Hadjod stem extract may be attributed to the presence of these bioactive compounds.

V. CONCLUSION

In conclusion, the Hadjod plant (Cissus quadrangularis) is a valuable medicinal herb with a long history of use in traditional medicine, especially for healing bone fractures and improving joint health. Modern research supports many of these traditional claims, showing that the plant has strong antioxidant, anti-inflammatory, and bone-healing properties.

The presence of important phytochemicals like flavonoids, triterpenoids, and vitamin C contributes to its therapeutic effects. These compounds help reduce inflammation, promote faster healing of bones, and protect the body from oxidative stress. Because of these benefits, Hadjod has gained attention as a potential natural alternative for managing conditions like osteoporosis and arthritis.

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