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The Pharma Innovation



ISSN (E): 2277-7695 ISSN (P): 2349-8242 NAAS Rating: 5.23 TPI 2022; SP-11(6): 808-810 © 2022 TPI www.thepharmajournal.com Received: 12-03-2022

Accepted: 15-04-2022

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Preliminary phytochemical analysis of aqueous and alcoholic extracts of *Moringa oleifera* leaves

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Abstract

Phytochemical analysis is very important tool for identifying the quality as well as medicinal importance of particular plant. *Moringa oleifera* (Sainjna, Drumstick tree) an ancient plant described in Vedas, Sushruta Samhita and in various nighantu is a very important medicinal plant. Ethno-pharmacologically, Moringa is used in many diseases *viz*. Helminthiasis, wound healing, diabetes, cancer, inflammation and various other disease conditions. Present study was carried out to find the phytochemical constituents of various extracts of *Moringa oleifera* leaves and its anthelmintic activity. Dried leaves of Moringa were taken and then grinded to make coarse powder of it, later its aqueous and alcoholic extracts prepared in Soxhlet apparatus. Phytochemical analysis of aqueous and alcoholic extracts of *Moringa oleifera* leaves revealed presence of alkaloids, reducing sugars, glycosides, proteins, tannins, flavonoids and saponins.

Keywords: Moringa oleifera, helminthiasis, phytochemical, flavonoids, leaves

Introduction

The phytochemical studies were performed/ used for testing the different chemical groups present in the drug or extracts (Breslin Andrew, 2017)^[3]. It defines the screening, extraction and identification of the medicinally principal active ingredients in the plant. These are produced either by primary or secondary metabolism (Molyneux, 2007) ^[7]. The secondary metabolites are naturally occurring in various parts of plants like leaves, vegetables and roots that have defense mechanism and protect from various disease (Baruah et al., 2018)^[2]. Phytochemicals are non-nutritive, chemical compounds and occur naturally on plants during metabolic processes having diverse defensive actions or disease preventive properties. Plants are known to produce these chemicals to protect them (Minakshi, et al., 2016) [6]. Phytochemicals appear to neutralize free radicals, inhibit enzymes that activate carcinogens and activate enzymes that detoxify carcinogens (Saxena, 2013) [10]. Moringa oleifera can prevent more than 300 diseases and is considered as one of the most useful plant as almost every part can be used for food or has some other advantageous properties. In the tropics, it is used as forage for livestock and in other countries also used to treat various ailments (Ganguly, 2014; Tayo et al., 2014) [4, 11]. Leaves of Moringa oleifera are commonly used and have various bioactive compounds, which are responsible for pharmacological activities like, anthelmintic, anti-inflammatory, hypolipidemic and anti-diabetic etc. (Konmy et al., 2016) ^[5]. The present study was aimed to evaluate Qualitative phytochemical analysis of aqueous and alcoholic extracts of Moringa oleifera leaves.

Materials and Methods

The phytochemical analysis was undertaken to determine the presence of various active constituents of aqueous and alcoholic extracts of *Moringa oleifera* leaves by conducting the various tests described by Raman, 2006^[9].

1. Test for alkaloids

A little extract was taken in 5 ml of 1.5% hydrochloric acid (v/v) and filtered through Whitman's filter paper No. 1. The filtrate was used for testing alkaloids.

A. Dragendorff's test: Dragendorff's reagent: It was prepared by mixing solution a (1.7 g of bismuth sub nitrate + 20 g of tartaric acid + 80 ml of distilled water) and solution B (16 g potassium iodide + 40 ml of distilled water) in equal (1:1) proportion (v/v) and allowed to

stand for a few minutes. From this solution a working standard was prepared by taking 10 ml of this solution and adding 20g of tartaric acid and making its volume up to 100 ml with distilled water.

Procedure: The working reagent was sprayed on a filter paper and the paper was dried. The sample solution was applied on the paper using a capillary tube. Development of an orange-red colour indicates the presence of alkaloids.

B. Wagner's test: Wagner's reagent: 1.27 g iodine and 2.0 g potassium iodide were dissolved in distilled water and the total volume was made to 100 ml for preparing Wagner's reagent.

Procedure: Take 5ml of acid solution of the extract and then added 2ml of Wagner's reagent in it. Appearance of brown flocculent precipitate indicates the presence of alkaloids.

3. Test for reducing sugars

- **A. Benedict's test:** About 5 ml of dissolved extract was taken with equal quantity of Benedict's reagent and heated. The appearance of brownish red precipitate (reduction) was indicative to presence of reducing sugars.
- **B.** Fehling's test: Two ml of aqueous solution of extract in a test tube was added into 5 ml of Fehling's reagent (mixture of equal volumes of Fehling's solutions A and B) and boiled in a water bath for about 2 min. The brickred precipitate was indicative of the presence of reducing sugars.

4. Test for glycosides

- **A. Benedict's reagent test:** The solution obtained in Benedict's test for reducing sugars was filtered and 1 ml dilute hydrochloric acid was added to it for hydrolyzing the glycosides. Equal quantity of Benedict's solution was added to it and boiled in hot water bath. Appearance of brownish precipitate suggested the presence of glycosides.
- **B.** Fehling's reagent test: This test is performed with the solution obtained in Fehling's test. To the clear solution a few drops of dilute hydrochloric acid was added and boiled for 5 minutes for hydrolyzing glycosides. Fehling's reagent was again added to note any further reduction, which indicates the presence of glycosides.

5. Test for sterols

- A. Salkowski reaction: 0.5 g of extract was dissolved in 2 ml concentrated chloroform in a test tube. Then 2ml of concentrated sulphuric acid was added to it by the side wall of test tube slowly drop by drop. Development of red colour in the chloroform layer and greenish florescence in the lower part of solution are suggestive of the presence of sterols in extracts.
- **B.** Lieberman Buchar reaction: 0.5 g of extract was dissolved in 2 ml chloroform in a test tube. 5-10 drops of acetic anhydride were added followed by addition of 0.5 ml concentrated sulphuric acid by the side wall of the test tube. Transient colour development from red to blue and finally green colour indicates the presence of sterols.

6. Test for proteins

A. Xanthoprotein test: One gram of extract was taken in a test tube containing 2 ml distilled water and 0.5 ml

concentrated nitric acid was added to it. Appearance of a yellow precipitate suggests the presence of protein.

B. Biuret test: One gram of extract was taken in tube having 1 ml distilled water. Then 1 ml of 1 per cent sodium hydroxide solution was added to it. To this, 1-2 drops of 1 per cent copper Sulphates solution were added. Development of violet colour indicates the presence of protein.

7. Test for tannins

One gram of extract was taken with 5 ml of methanol and warmed and then filtered. The filtrate was divided into two parts and tested with following reagents.

- **A. Lead acetate test:** A few drops of lead acetate solution were added to the methanolic extract. The formation of precipitate indicates the presence of tannins.
- **B.** Ferric chloride test: A few drops of ferric chloride solution were added to the little of the above filtrate. Development of green colour in the filtrate indicates the presence of tannins.

8. Test for flavonoids

- **A.** Alkaline reagent test: Extract was treated with few drops of sodium hydroxide solution. Formation of intense yellow colour, which becomes colorless on addition of dilute acid, indicates the presence of flavonoids.
- **B.** Lead acetate test: Extract was treated with few drops of lead acetate solution. Formation of a yellow colored precipitate indicates the presence of Flavonoids.

9. Test for saponins

Foam test: One gram of extract was taken in a test tube containing 5 ml of sodium bicarbonate and 5 ml of water and, the contents were shaken vigorously. Formation of a stable froth suggests the presence of saponins.

9. Test for Anthraquinones

Bontrager's Test: 0.5g of extract was boiled in a test tube for 1-2 minutes with 5 ml of 10 per cent sulphuric acid and filtered immediately. The filtrate was cooled and shaken with benzene. The benzene layer was separated and the same was shaken with half of its volume of 10 per cent ammonia. Development of pink ring in ammonical layer indicates the presence of Anthraquinones.

Results and Discussion

Aqueous and alcoholic extracts of Moringa oleifera leaves gave the presence of following phytochemicals are presented in Table 1. In the present study, aqueous extract of Moringa oleifera leaves revealed the presence of alkaloids, reducing sugars, glycosides, sterols, proteins, tannins, flavonoids and saponins in the present study. Adline and Devi (2014)^[1] reported the presence of tannins, glycosides, flavonoids and terpenoids in aqueous extract of Moringa oleifera leaves. Alcoholic extract of Moringa oleifera leaves revealed the presence of alkaloids, reducing sugars, glycosides, proteins, tannins, flavonoids and saponins. Adline and Devi (2014)^[1] reported the presence of tannins, glycosides, flavonoids, terpenoids and phenol in ethanolic extract of Moringa oleifera leaves. Patel et al. (2014)^[8] showed the presence of flavonoids, tannins, steroids, alkaloids, saponins in ethanolic and aqueous extract of Moringa oleifera leaves on phytochemical screening. The phytochemical investigation of ethanolic and aqueous extracts of Moringa oleifera leaves revealed the presence of various active principles like tannins, alkaloids, steroids, triterpenoids, flavonoids, hydroxy-

Anthraquinones, cardiac glycosides, and carbohydrates (UC and Nair, 2013) $^{[12]}$.

Table 1: Qualitative phytochemica	al analysis of aqueous and alcohol	lic extracts of Moringa oleifera leaves.
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Active principle	Test applied	Aqueous extracts of Moringa oleifera leaves	Alcoholic extracts of Moringa oleifera leaves
Alkaloids	Dragendorff's reagent	+	+
	Wagner's reagent	+	+
Reducing sugars	Benedict's reagent	+	+
	Fehling's reagent	+	+
Glycosides	Benedict's reagent	+	+
	Fehling's reagent	+	+
Sterols	Salkowski reaction	+	-
	Lieberman Buchar reaction	+	-
Proteins	Xantho protein test	+	+
	Biuret test	+	+
Tannins	Lead acetate test	+	+
	Ferric chloride test	+	+
Flavonoids	Alkaline reagent test	+	+
	Lead acetate test	+	+
Saponins	Foam test	+	+
Anthraquinones	Bontrager's test	-	_

Key: + = Present; - = Absent;

Conclusion

Qualitative phytochemical analysis of both aqueous and alcoholic extracts revealed presence of alkaloids, reducing sugars, glycosides, proteins, tannins, flavonoids, proteins and saponins. Sterols were present in aqueous extract of *Moringa oleifera* leaves only. So we can say that various active principles which revealed during phytochemical analysis of both extracts of *Moringa oleifera* leaves which help in anthelmintic activity. So it reveals that *Moringa oleifera* is very important medicinal plant with various therapeutic applications.

Acknowledgement

Authors are thankful to the Prof. A.P. Singh, Principal Investigator, and Dr. Ashok Gaur, Asst. Professor and Co-P.I., Centre for Ethno Veterinary Practice and Alternative Medicine (CEVPAM), RAJUVAS, Bikaner for providing laboratory facilities in this research. Authors are also thankful to the Dr. Suchi Chatterjee, Joint director, Regional Disease Diagnostic Laboratory (RDDL), Animal Husbandry Deptt., Govt. of Rajasthan, Bikaner, for providing laboratory facilities for hematological estimations conducted at RDDL.

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