International Journal of Botany Studies
www.botanyjournals.com
ISSN: 2455-541X
Received: 30-06-2021, Accepted: 15-07-2021, Published: 30-07-2021
Volume 6, Issue 4, 2021, Page No. 481-485

Phytochemical study, antioxidant and anti-inflammatory activity of the *Euphorbia mauritanica*

Nitin A Sasane[1], Paresh S Gaikar[2], Arun Chandore[3], Kisan Pathade[4], Gurumeet C Wadhawa[2*]
[1] Mahatma Phule Arts, Science and Commerce College, Panvel, Maharashtra, India
[2] Rayat Shikshan Sanstha's, Karmaveer Bhaurao Patil College, Vashi, Navi Mumbai, Maharashtra, India
[3] Rayat Shikshan Sanstha's Abasaheb Marathe Arts and New Commerce, Science, College, Rajapur (VG), Maharashtra, India
[4] Rayat Shikshan Sanstha's, Maharaja Jivajirao Shinde Mahavidyalaya, Shrigonda, Maharashtra, India

Abstract
Euphorbia mauritanica is the name of a plant in the Euphorbiaceous family. This is a rare plant with high activity that has been used to treat a wide range of ailments and infections. In present study, flowers and fruits of *Euphorbia mauritanica* plant is used for phytochemical analysis and biological activities such as antioxidants and anti-inflammatory activities. In phytochemical analysis, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*. sterols, tannins, phenolic nucleus, terpenes, reducing sugars, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*. Techniques for determining antioxidant and anti-inflammatory activities that are well-known. Both the flowers and fruit sex tract of *Euphorbia mauritanica* have substantial antioxidant and anti-inflammatory properties. Anti-inflammatory activity is maximum in ethanol extract for flowers and fruits extracts of *Euphorbia mauritanica*.

Keywords: antioxidant, anti-inflammatory, fruits, flowers, *Euphorbia mauritanica*, phytochemicals

Introduction
Euphorbia mauritanica is a monoecious, spineless, many-branched succulent shrub. It is about 1.5 m high, with branches arising from a thick root stock. *Euphorbia mauritanica* grows comfortably in dry climates, inland areas or colder winters. The branches of *Euphorbia mauritanica* are pencil-thin, yellowish green, cylindrical and smooth, with alternate leaf scars. *Euphorbia mauritanica* is reputed to be venomous and only steenbuck and klipspringer are known to eat it [1]. The plantscantain various phytochemical and that helpful for the biological activity against diseases. Phytochemicals like saponins, flavonoids, terpenesetc. are naturally occurring compounds in plants that provide colour, flavour and aroma to fruit and vegetables [2]. Phytochemicals are also known as phytonutrients. They are biologically active [3]. Phytochemicals shows antioxidants, anti-inflammatory, antibacterial properties, which are helpful to lower the risk of chronic diseases in body. Antioxidants are compounds that protect body against toxic effect of free radicals. Inhibit oxidation and protect cell from damage caused by unstable molecule generating process of oxidation during metabolism in body such as free radicals [4]. Free radicals lead to chain reaction and start damaging cells. When free radical level in body become high, it creates oxidative stress [5]. Oxidative stress plays important role in variety of diseases like cardiovascular problems, cancers, diabetes. Lipid's proteins nucleic acids are major targets of free radicals [5]. The protective mechanism of antioxidants serves to scavenge free radical and prevent initiation of chain reaction. For proper physiological functions balance between antioxidants and free radicals is necessary [6]. Inflammation is critical protective response of body infection, irritation injury, Swelling, heat, redness, pain are some symptoms of inflammation [6]. Cell death, tissue injury, ischemia, cancer arealso responsible for inflammation. Inflammation can be acute or chronic. Chronic inflammation last long. Anti-inflammatory compounds are usedto treat symptoms of inflammation in body [8]. In the present study, various phytochemicals detection analysis caried with standard protocol of flowers and fruits of *Euphorbia mauritanica* plant. In addition, for various extract antioxidant and anti-inflammatory activity is examined of *Euphorbia mauritanica* plant flowers and fruits.

Fig 1: *Euphorbia mauritanica* Plant

Materials and Methods

The flowers and fruits of *Euphorbia mauritanica* plant were collected from Lonavala, Maharashtra. The specimen of fruits and flowers washed with distilled water then specimens are dried. After completion of drying, then pulverized the specimens and powered of fruits and flowers *Euphorbia mauritanica* plant stored in air tight bottles. The flower and fruits extract of the *Euphorbia mauritanica* plant were named EM-F and EM-Fr respectively.

Phytochemical Analysis

Phytochemical testing carried using estimated protocol for EM-F and EM-Fr extract [9].

Sterols

An equal volume of acetic anhydride was added to test tube and gently stirred. After that, 1 ml of concentrated H_2SO_4 was poured down the tube's side [10]. The presence of sterols is indicated by the formation of a brownish-red ring at the contact zone of the two liquids and a greenish tint in the separation layer. Test is Positive for both EM-F and EM-Fr.

Tannins

Ferric chloride test was performed for detection of tannins [11]. In this test, appearance of a blue changed to olive green as additional ferric chloride was added. Ferric chloride test is positive for EM-F and EM-Fr.

Anthracene

5 mL chloroform was added to the powder of flowers or fruits in a test tube and agitated for 5 minutes. The mixture was filtered, and the filtrate was agitated with a 10% ammonia solution in an equal volume. When the aqueous layer is agitated, it turns pink, red, or violet, indicates the presence of free anthraquinone [12]. Anthracene detection test was positive for the EM-F and negative for EM-Fr.

Saponins

The powdered flowers or fruits were put to a test tube with 10 mL distilled water and vigorously shaken for 30 seconds. Afterwards, it was left to stand for 30 minutes. The presence of saponins is indicates by the production of honeycomb foam [6]. Saponins's detection test was Positive for EM-Fas well as negative for EM-Fr.

Flavonoids

Acetone was used to totally retain two grams of powder of flowers or fruits. After evaporating the acetone over a water bath, the residue was removed with warm water. After filtering the mixture while it was still hot, the filtrate was allowed to cool before being used for the next test: Shinoda's experiment in 3 ml of an aqueous solution, a few magnesium chips were added, and 2 drops of weak HCl were added and warmed. The presence of flavonoids is indicates by a pink or red tint [13]. Flavonoid's detection test was Positive for EM-Fas well as negative for EM-Fr.

Phenolic nucleus

Sodium hydroxide test is used for detection of phenolic nucleus [14]. Phenolic nucleus's detection test was Positive for EM-Fas well as negative for EM-Fr.

Terpenes

The Liebermannre agent test aids in the identification of terpene, resulting in the production of a blue green colour that shows the presence of a terpene, whereas no pink colour shows the absence of terpenes [15]. Terpene's detection test was Positive for EM-Fas well as negative for EM-Fr.

Reducing Sugars

The Fehling reagent was used to identify reducing sugars, which was then validated by the Tollens reagent test [13]. Reducing Sugar's detection test was Positive for EM-Fand EM-Fr.

Alkaloids

Bouchardat reagent and (reagent iodo-iodized) Bouchardat reagent were used to characterize alkaloids [16]. Alkaloid's detection test was Positive for EM-Fas well as negative for EM-Fr.

Proteins

The biuret reaction was used to detection the proteins. Add 2 -3 drops of an aqueous portion of CuSO4 to 2% to a small volume of extract diluted in 2 mL of 20% aqueous NaOH in a test tube. Purple colour formations indicates the presence of protein [17]. Proteins's detection test was Positive for EM-Fas well as negative for EM-Fr.

Coumarins

2 mL ethanolic solution produced from each residue during extraction in two test tubes Heats both test tubes in a water bath until boiling, then add 0.5 mL of 10% NaOH to one of the test tubes. 4 mL distilled water in each test tube to cool it down. If the liquid from the test tube in which the alkaline solution was added is transparent or more translucent than the liquid from the control test tube (without the alkaline solution), a faint yellow solution indicates the presence of coumarin [3]. Coumarin's detection test was positive for EM-Fas well as negative for EM-Fr.

Saponosides

8-10 mL aqueous complete extract in a test tube to discover saponosides. The tube was shaken for 10-15 seconds before being left alone for 12-15 minutes. saponosidessaponins are detects when the height of persistent foam was greater than 1 to 2 cm [14]. saponoside's test was positive for EM-Fas well as negative for EM-Fr.

Antioxidant Activity Determination

DPPH Scavenging Test: The percentage of the antioxidant present in the sample was determined using the typical protocol of the DPPH scavenging test. This test was carried out using the specific protocol. This test was carried out by preparing the various extracts of the plant material [18].

Study of anti-inflammatory activity (In-vitro models)

The anti-inflammatory activity of the different extracts was carried out using a slight modification of Mizushima and Kobayashi protocol with doses. The albumin test method was used [19].

Results and Discussions

Phytochemical Analysis

Based on the present study it can be concluded that the ethanolic extract from fruits and flowers *Euphorbia mauritanica* showed the presence of various phytochemicals. In phytochemical analysis, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*. Sterols, tannins, phenolic nucleus, terpenes, reducing sugars, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*.

Antioxidant Activity Determination

The antioxidant activities of the organic solvents of flowers and fruits extract of *Euphorbia mauritanica* tabulated in table 1 and table 2 for BHT, C_2H_5OH, $CHCl_3$ and CCl_4 extract. The graphical performance of BHT, C_2H_5OH, $CHCl_3$ and CCl_4 for EM-F displayed in Fig. 2 (a). It shown that C_2H_5OH and $CHCl_3$have better antioxidants performance than CCl_4. Fig. 2 (b) displayed that DPPH radical activity of EM-Fr and $CHCl_3$ and CCl_4 exhibited better performance than C_2H_5OH extract.

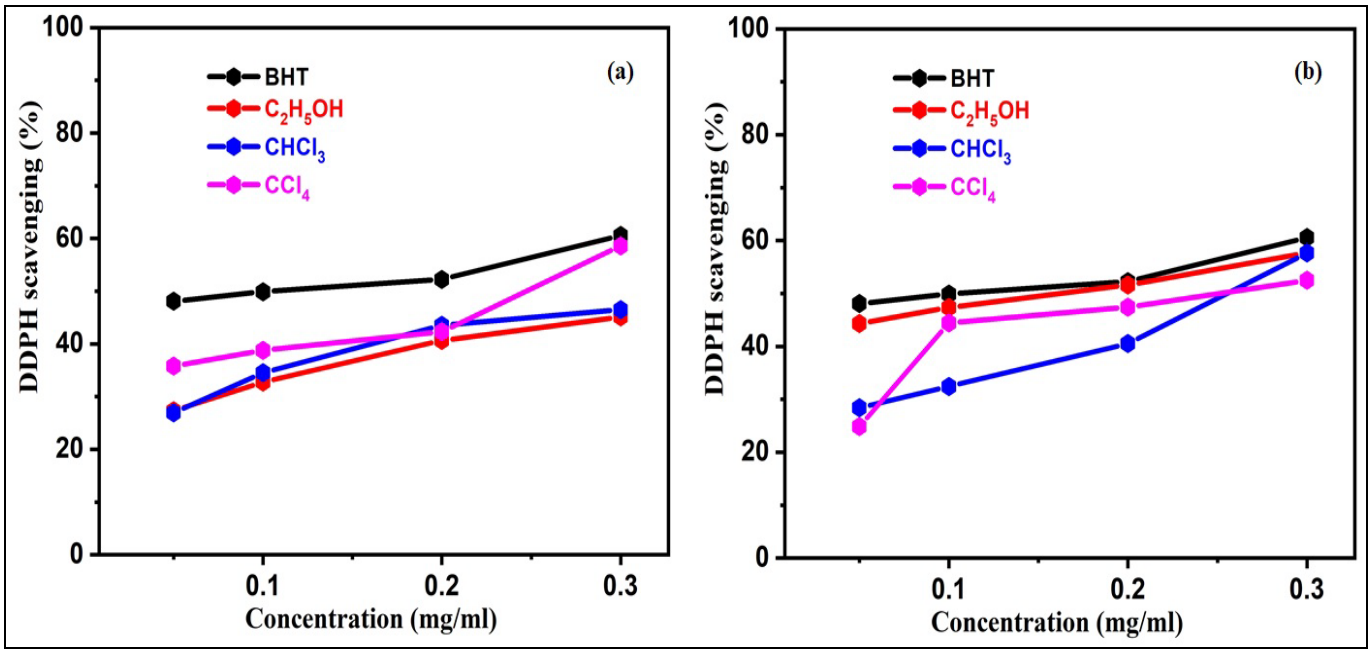

Fig 2: Antioxidant activity of EM-F and EM-Fr a) DPPH radical activity of EM-F b) DPPH radical activity of EM-Fr

Table 1: Antioxidant activity of EM-F

Extract Conc. Mg/ml	BHT	Ethanol	$CHCl_3$	CCl_4
0.05	48.1	27.30	26.93	35.80
0.1	49.91	32.77	34.53	83.79
0.2	52.24	40.66	43.54	42.32
0.3	60.57	45.12	46.50	58.60

Table 2: Antioxidant activity of EM-Fr

Extract Conc. Mg/ml	BHT	Ethanol	$CHCl_3$	CCl_4
0.05	48.1	44.34	28.44	24.88
0.1	49.91	47.34	32.44	44.45
0.2	52.24	51.63	40.53	47.40
0.3	60.57	57.66	57.67	52.50

Determination of Anti-inflammatory Activity

Anti-inflammatory activity (*In-vitro* models) studiedfor *Euphorbia mauritanica* plant's flowers and fruits extract tabulated in table 3 and table 4 respectively for standard (Ibuprofen), petroleum ether, chloroform, ethyl acetate, n-Butanol and ethanol. Fig. 3 exhibited percent inhibition of EM-F and EM-Fr for standard, Petroleum Ether, Chloroform, Ethyl acetate, n-Butanol and Ethanol. Fig. 3 shown that anti-inflammatory activity is maximum in ethanol extract for both EM-F and EM-Fr than other extract. Mostly Ethanol extract of the flowers and fruits of *Euphorbia mauritanica* possess *in-vitro* anti-inflammatory activity which might be attributed to the presence of various phytochemicals in the extract. The chloroformextract found less performance in EM-Fr while ethyl acetate extract found less performance in EM-F.

Table 3: Anti-inflammatory activity of EM-F

In-Vitro Anti – inflammatory activity	Dose (mg / kg)	Absorbance value (Mean + SE)	Inhibition of denaturation (%)
Control	5ml / kg	0.098	----
Standard (Ibuprofen)	100mg/kg	0.18	90.32
Petroleum ether extract	200mg/kg	0.15	57.40
Chloroform extract	200mg/kg	0.14	56.25
Ethyl acetate extract	200mg/kg	0.12	43.26
n-Butanol	200mg/kg	0.16	54.37
Ethanol	200mg/kg	0.17	78.47

Table 4: Anti-inflammatory activity of EM-Fr

In-Vitro Anti – inflammatory activity	Dose (mg / kg)	Absorbance value (Mean + SE)	Inhibition of denaturation (%)
Control	5ml / kg	0.098	----
Standard (Ibuprofen)	100mg/kg	0.18	90.32
Petroleum ether extract	200mg/kg	0.12	68.19
Chloroform extract	200mg/kg	0.13	24.20
Ethyl acetate extract	200mg/kg	0.12	54.18
n-Butanol	200mg/kg	0.14	67.71
Ethanol	200mg/kg	0.12	85.18

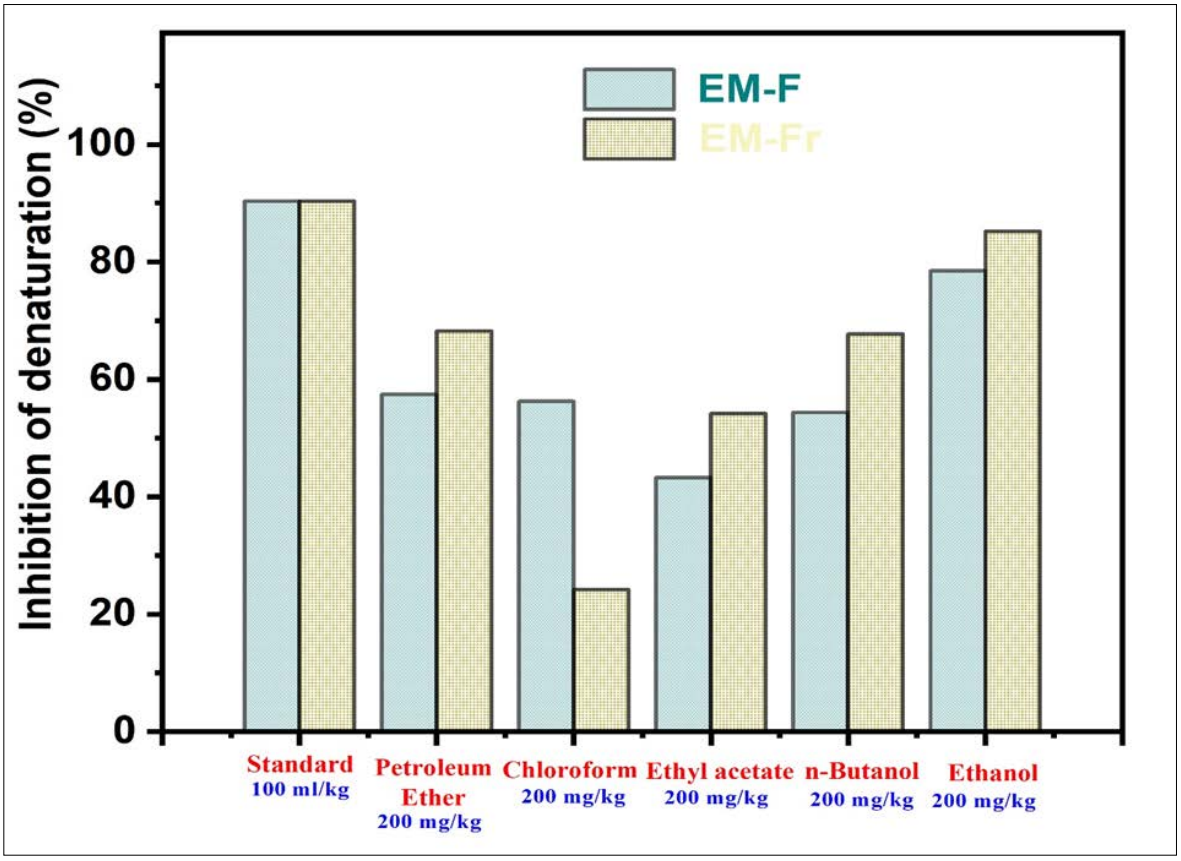

Fig 3: % Inhibition of EM-F and EM-Fr for standard, Petroleum Ether, Chloroform, Ethyl acetate, n-Butanol, Ethanol

Conclusions

In phytochemical analysis, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*. Sterols, tannins, phenolic nucleus, terpenes, reducing sugars, coumarins, saponosides were present in flowers extract of *Euphorbia mauritanica*. anti-inflammatory activity is maximum in ethanol extract for both fruits and flower extract. Fruits and flowers of *Euphorbia mauritanica* strong antioxidant activity as evidenced by the free radical scavenging property, can be a very effective antioxidant and can protect against the oxidative stress that is found to be an important pathophysiological event in a variety of diseases including aging, diabetes, cancer, cardiovascular disorders, and rheumatoid arthritis. Overall, itis a source of natural antioxidant that can be important in disease prevention and health preservation. Therefore, its ethnomedical claims was true according to the above experimental results. This gives support to the claim for the traditional use of the plant in the treatment of inflammation. The result of the study has seen to provide support for the use fruits and flowers of *Euphorbia mauritanica* to promote proper conservation sustainable use of such plant resources, awareness of local communities should be enhanced incorporating the traditional knowledge with scientific findings.

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