Sana'a University Journal of Applied Sciences and Technology

Vol. 1 | No. 1 | Page 13 – 18 | 2023 | https://jpurnals.su.edu.ye/jast

Antioxidant activity, total phenol contents, and antimicrobial activity of two Yemeni medicinal plants belongs to Euphorbiaceae family (*Jatropha curcas* and *Euphorbia Cactus Ehrenb*

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ARTICLE INFO
Article history:
Received: January 15, 2023
Accepted: January 31, 2023
Published: January, 2023

Keywords
1. Jatropha curcas
2. Euphorbia Cactus Ehrenb
3. Antioxidant activity
4. total phenol contents
5. antimicrobial

ABSTRACT: Jatropha curcas and *Euphorbia Cactus Ehrenb* plants, which belong to the Euphorbiaceae family are employed to cure different infections in traditional folklore medicine. Antioxidant activity and total phenol contents (TPCs) for latex of these two plants were analyzed and examined its antimicrobial effect. Both plants showed high antioxidant activity and highly phenolic contents. A positive correlation between antioxidant activity and total phenols has been noted. *Jatropha curcas* latex demonstrated significant inhibition against the test microorganisms. These plants were demonstrated as important for reconnoitering are new biomolecules that will be used in new pharmaceuticals.

CONTENTS

1.Introduction

Plant extracts are a source of a large number of biologically active compounds. Natural products from various plant sources can have notable medicinal value and can be used for treatment different diseases. Researchers have tried to extract the most potent antimicrobial agents from different types of sources, like plants used in conventional folk medicines [1], [2]. The use of medicinal plants as traditional medicine in most developing countries, as a basis for the upkeep of good health, has been broadly observed and approximately 80% of the world's inhabitants depend on herbal medicine[2], [3]. Presently, Top pharmaceutical companies are inaugurating their herbal plants to placate the blossoming exigency of herbal medicines in their countries[4].

Euphorbiaceae is a family that includes around 300 kinds of plants, being one of the largest and most ecumenical families in angiosperms sub-branching [5].

The largest genus in the plant family Euphorbiaceae is the genus Euphorbia, which includes circa 2000 noted species and ranges from trees to annuals. All have unique flower structures and contain latex [6], [7]. *Jatropha curcas* and *E. cactus Ehrenb* are medicinal plants belonging to the Euphorbiaceae family.

Jatropha curcas is a plant species reported from the southern Arabian Peninsula and central Africa that variously known as Barbados nut and known locally in Yemen as Yatrophat Al-Gahta. It is a small tree about 5 m high. various skin diseases such as wound healing [8], [9], rheumatism, and coughs can be treated using *Jatropha curcas* latex. It has also been used to treat various diseases caused by pathogenic microbes [10]–[12]. The latex of *Jatropha curcas*'s stem contains hydrocyanic acid, tannins (10%), jatrophine, and toxalbumine. In many countries, the fruit/seed of *Jatropha curcas* is used as biofuel because it is riches in fat/oil, which also contains phytic acid (12%), trypsin inhibitors, curcin, and saponins [13].

E. cactus Ehrenb is a succulent spiny leafless shrub, with about 1 m high and 2-4 flowers in the branches of the upper part of the plant. Seeds are brownish-grey with 2.7 mm diameter. This species distributed in the South Arabian Peninsula and Africa (Sudan and Eritrea). The latex of E. cactus Ehrenb contains bioactive compounds which have anti-leishmanial activity, that could ministrate as an alternative agent in the treatment of Cutaneous leishmaniasis [7].

Euphorbia Cactus Ehrenb

Jatropha curcas

The most of research studied the pharmacology of leaves, roots, and steam extract for *Jatropha curcas* and *Euphorbia Cactus Ehrenb* plants. No more researches studied the latex, therefor the aim of our study to evaluate the antioxidant activity, antimicrobial activity, and total phenol content for *Jatropha curcas* and *Euphorbia Cactus Ehrenb* latex. Additionally, the present work showed that a correlation exists between TPCs content and (FBRC and FRAP). Such research is important for reconnoitering new biomolecules that will be used new pharmaceuticals.

2. Material and Methods

2.1 Collection of Plant Material

The current study was carried out in the Department of Chemistry, Faculty of Science, Sana'a University, Sana'a, Yemen from November, 2021. The target plant, *E. cactus* Ehrenb, was collected from Atma reserve in the Atma district of Dhamar Governorate, Yemen and was identified by taxonomist Dr.Hassan M. Ibrahim, head of the

Herbarium, Biological Science Department, Faculty of Science, Sana'a University, Yemen.

2.2 Preparation of Samples

The latex part of the plant was dissolved in dimethyl sulfoxide (DMSO) (0.1 mL/ mL) for both antioxidant and antimicrobial samples.

2.3 Ferric-Bipyridine Assay (FBRC)

This method is based on the formation of the ferrous form of the Fe^{3+}-Bipyridine complex through reduction at low pH = 3.6. Measurement of antioxidant activity by monitoring the change in absorption has been taken at 535 nm. Ferric-Bipyridine solution consists of 2 mL of

(2 mL) then the volume was made up to 10 mL. Measurement of absorbance were taken at 610 nm with a spectrophotometer and relating to ascorbic acid standards. The results of antioxidant capacity were shown as (mg ascorbic acid equivalents /L of latex) [15].

2.4 Ferric Reducing Antioxidant Power Assay (FRAP)

This method is based on the formation of the ferrous form of the Fe^{3+}-TPTZ complex through reduction at low pH. Measurement of antioxidant activity by monitoring the change in absorption has been taken at 610 nm. FRAP solution consists of CH_3COOH/CH_3COONa buffer (300 mmol/L) at pH = 3.6, TPTZ (9.9 mmol/L) and $FeCl_3.6H_2O$ (10.07 mmol/L) (10 v /1 v/ 1 v). 50 µL of the plant latex dissolved in DMSO (0.01 mL/mL) was added to the FRAP solution (2 mL) then the volume was made up to 10 mL. Blank solution was made of FRAP solution

2.5 Total Phenol Content (TPC)

This method reducees yellow heteropolyphosphomolibdatetungstate anions to a blue color which determines phenols and oxidized substances. 50 µL of the plant latex dissolved in DMSO (0.01 mL/ mL) was added to 1 mL of a 1:10 diluted Folin and Ciocalteu reagent. After 5 min, 1 mL of a Na_2CO_3 solution (7.5 % w/v) was added. The volume was made up to 10 mL with deionized water. Measurements of the absorbance for solution were taken at 760 nm against water blank. A standard curve has been prepared with 2-20 µmol/L CH_3COOH/CH_3COONa buffer (300 mmol/L) at pH = 3.6, 1 mL of B.p (6.5 mmol/L) and 1 mL of $FeCl_3.6H_2O$ (10.07 mmol/L) . 50 µL of the plant latex dissolved in DMSO (0.01 mL/1 mL) was added then the volume was made up to 10 mL. Blank solution consists of 2 mL of buffer solution (300 mmol/L) at pH = 3.6, 1 mL of B.p (6.5 mmol/L) and 1 mL of $FeCl_3.6H_2O$ (10.07mmol/L) then the volume was made up to 10 mL. Measurement of absorbance were taken at 535 nm with a spectrophotometer relating to ascorbic acid standards. The result of antioxidant power was shown as mg ascorbic acid equivalents / L of latex [14].

Ascorbic acid and was compared to the optical density (OD) of the sample. Results were shown as ascorbic acid equivalents (AAE) [16].

2.6 Percentage of Phenols to total antioxidants

The percentage of phenols and oxidized substances was determined through dividing the concentration of polyphenolic compounds determined through TPC method by the antioxidants concentration determined through FRAP and FBRC methods then multiplying the results by 100 according to the following equations:

a) Phenols and Oxidized Substances % = (TPCs/FRAP) x 100

b) Phenols and Oxidized Substances % = (TPCs/FBRC) x 100

Percentage values were taken as a range between the results given through equation of

ascorbic acid equivalents (AAE) [17].

3. Result and Discussion

3.1 Antioxidant Activity and Total Phenol Contents:

Newly, synthetic food antioxidants might be substituted by plant extracts especially that contained polyphenolics, which may impact human health when consumed constantly [18].

The antioxidants activity using (FBRC and FRAP) methods and total phenolic content (TPC) through analysis of Folin-Ciocalteau method of *E. cactus Ehrenb* and *Jatropha curcas* were determined (Table 1).

Table 1. FBRC, FRAP, and TPCs values express as (mg AAE L^{-1} latex) *E. cactus Ehrenb* and *Jatropha curcas*

species	FBRC (mgAAE/ L of latex)	FRAP (mgAAE/L of latex)	TPCs (mgAAE/L of latex)
E.cactus Ehrenb	15.18 ± 0.1	21.755 ± 0.1	2.49 ± 8.4E-2
Jatropha curcas	420.72 ± 0.1	293.97 ± 0.1	183.38 ± 0.57

The measurements of antioxidant activity via FBRC and FRAP methods and total phenol content indicated that the both *Jatropha curcas* and *E. cactus Ehrenb* have antioxidant capacities and TPCs, therefor *Jatropha curcas* has higher antioxidant and total phenol contents than *E. cactus Ehrenb*. *Jatropha curcas* antioxidant capacity is very tremendous that can be considered the latex of *Jatropha curcas* has valued antioxidant agents which accomplish the pharmaceutical needed.

3.2 Correlation between antioxidant activity and total phenol contents

The regression equation and the correlation coefficient (r^2) between TPCs and (FBRC and FRAP) were calculated using orgin software system. Therefor it has been a significant positive correlation between the TPCs present in the latex and its antioxidants.

The correlation with TPC contents was found to be high (R^2 = 1 and R^2 = 0.993,0.995Fig. 1) for *E. cactus Ehrenb* latex and *Jatropha curcas* respectively. That means that phenolic compounds are the main class of natural antioxidants in both *E. cactus Ehrenb whereas Jatropha curcas has less* R^2 that means that there are many other antioxidants contributing to overall antioxidant activity of latex [19].

3.3 The ratio of total phenol contents to total antioxidants

Table 2. showed that the percentage of TPCs to FBRC and FRAP are lowest for *E. cactus Ehrenb perhaps*, the most antioxidants are phenols as shown in Fig. 1, whereas the percentage of TPCs to FBRC and FRAP are the highest for *Jatropha curcas* therefor there are another antioxidant compounds beside polyphenols compounds (Fig. 1).

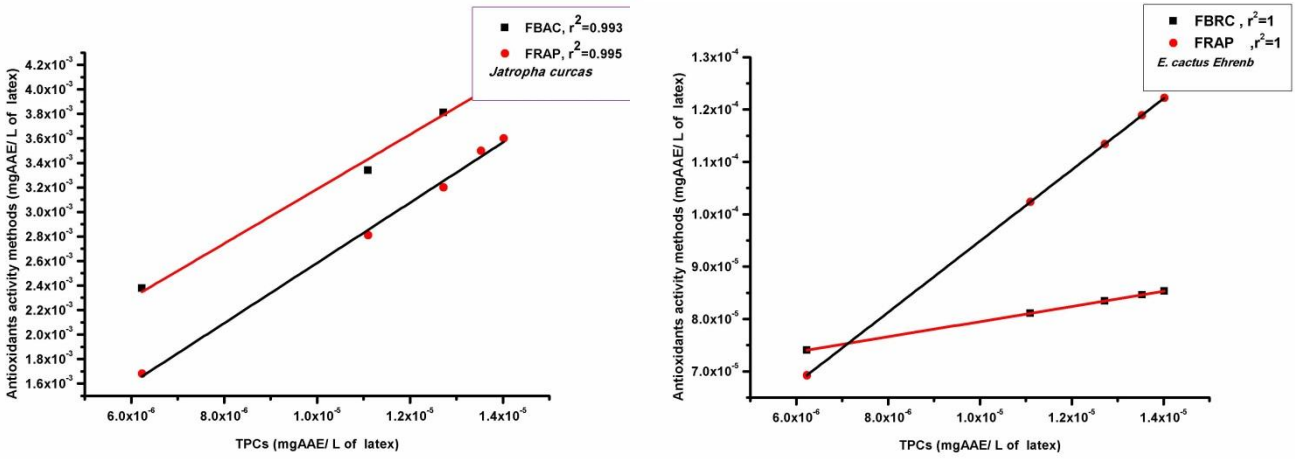

Fig.1 Linear correlation plot of (TPCs) as a function of (a) FBRC, and (b) FRAP

Table 2. The ratio of total phenol contents to total antioxidants for *E. cactus Ehrenb* and *Jatropha curcas*

Species	% TPCs/FBRC	% TPCs/FRAP
E. cactus Ehrenb	16.4	11.44
Jatropha curcas	43.6	62.36

3.4 Antimicrobial Activity

For last few years, scientists have been evaluating the antimicrobial properties by working on natural plant extracts for the development of new therapeutics[20]. The antimicrobial activity of *E. cactus Ehrenb* and *Jatropha curcas* were summarized in Table 3. *Jatropha curcas* latex demonstrated significant inhibition against the test microorganisms. The latex is active against Staphylococcus aureus. This agrees with the report of M. O. Oyama et al [21].

The *E. cactus Ehrenb* showed no antimicrobial activity at the concentrations tested. Since no antimicrobial investigation has been carried out on E. cactus Ehrenb. These results were agreed by Al-faifi, Zarraq Issa [7].

Table 3. Antimicrobial Activiy of Latex of *E. cactus Ehrenb* and *Jatropha curcas*

Method	Sample Volume (µL)	Microorganim	Species	Growth Medium	Incubation temperature (°C)	Incubation time (h)	*E. cactus Ehrenb*	*Jatropha curcas*
Disk diffusion method	20	Bacteria	Escherichia Coli	Mueller Hinton Agar (MHA)	35 ± 2	24-26	-	+
	20		Pseudomonas Aeruginosa		35 ± 2	24-26	-	+
	20		Staphylococcus Aureus		35 ± 2	24-26	-	+
	20		Streptococcus Pneumoniae		35 ± 2	24-26	-	+
	20	Yeast	Candida Albicans	Sabouraud Dextrose Agar (SDA)	35 ± 2	24-26	-	-

Both medicinal plants showed no antifungal activity.

Conclusion

Therefore, the present observations suggest that the Latex of *E. cactus Ehrenb* and *Jatropha curcas* is a rich source of antioxidant activity and total phenol contents. Jatropha curcas is a potential source of bioactive antimicrobial agents therefor it can be used as an important antimicrobial agent in new pharmaceuticals.

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