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Antibacterial, Antioxidant Activity and Phytochemical Analysis of *Euphorbia hirta* Linn.

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Authors' contributions

This work was carried out in collaboration between both authors. Author SJ collected the information, designed, analyzed the data and wrote the protocol. Author PD concerned in formatting, editing and drafting the manuscript critically and approved the final manuscript.

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ABSTRACT

Objective: Crude extracts were prepared from aerial parts of *Euphorbia hirta* Linn. in four solvents i.e. water, acetone, ethanol and petroleum ether. Extracts were investigated for their phytochemical, antibacterial and antioxidant properties.

Methods: Plant extracts were prepared by using Soxhlet apparatus. Antibacterial potential was assessed by disc diffusion method against six bacterial strains viz. *Klebsiella pneumoniae* (MTCC NO 109), *Staphylococcus aureus* (MTCC NO 96), *Pseudomonas aeruginosa* (MTCC NO 2453), *Mycobacterium smegmatis* (MTCC NO 992), *Bacillus subtilis* (MTCC NO 2057) and *Chromobacterium violaceum* (MTCC NO 2656). Minimum inhibitory concentration was determined by microbroth dilution method, only to those bacterial strains which showed a significant zone of inhibition by disc diffusion method. The extracts were also examined for the presence of various phytoconstituents, total phenolic and flavonoid content using standard methods. Free radical scavenging activity was assessed by DPPH method.

Results: The plant extracts showed a wide spectrum of inhibition against the test pathogens thus justifying the use of plant in traditional medicine. Acetone, aqueous and ethanol extracts exhibited stronger antibacterial activity as compared to the petroleum ether extract. The maximum activity recorded was 17.6 ± 0.57 mm with 200 mg/ml concentration of ethanol extract against

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Klebsiella pneumoniae. Petroleum ether extract was found to be least effective as it showed zone of inhibition only against *Chromobacterium violaceum*. Aqueous extract of the plant showed highest antioxidant activity and minimum IC50 value (105.39 µg/ml) followed by acetone (371.0 µg/ml), ethanol (504.92 µg/ml) and petroleum ether (645.12 µg/ml) extracts. Aqueous extract showed highest TPC value (8.2 mgGAE/g), followed by acetone (6.4 mgGAE/g) and ethanol (5.84 mgGAE/g). Similarly, highest TFC value (4.6 mgQE/g) was also observed against aqueous extract, followed by acetone (2.05 mgQE/g) and ethanol extract (1.82 mgQE/g). Petroleum ether extract exhibited minimum TFC value i.e. 0.79 mgQE/g.

Conclusions: The present in vitro study revealed that *E. hirta* is a rich source of bioactive compounds with significant antibacterial and antioxidant activity. However, further investigation with ethno-pharmacological approach is needed for better understanding of its utility in health care.

Keywords: Antibacterial activity; antioxidant potential; DPPH assay; disc diffusion method; plant extracts.

1. INTRODUCTION

Plants are well known source of bioactive compounds hence has been used in traditional medicine since ancient times. Not only medicinal plants, but weeds have also been proved to contain various bioactive constituents and used in conventional medications in many parts of the world [1-5]. This is particularly practiced in rural population where modern health care facilities are limited so people depend upon the local biodiversity for their well being. Phytocompounds are of significant value to mankind, not only due to their therapeutic potential and also being devoid of side effects often associated with the synthetic drugs [6-9]. This has generated lot of interest in the last few years to investigate plant material for new bioactive compounds as they are one of the basis for modern medicine. So the need of the hour is to accentuate the relevance of traditional remedies which constitute a major part of the health care system in the developing countries and is rapidly entering the therapeutics in the developed countries.

Although medicinal plants have been explored extensively for their bioactive compounds, not much work has been done on weeds as they not considered of much use to mankind. In the present investigation efforts have been made to explore the medicinal potential of *E. hirta* a wild herbaceous plant. Though it is native to Central America but it is found in all tropical countries, including India and grows abundantly in open waste spaces, grasslands, roadsides and pathways. The stem is slender; reddish in color and covered with yellowish bristly hairs especially in younger parts. Leaves are about 5 cm long, oppositely arranged, lanceolate and greenish or reddish underneath. The stem and

leaves produces white milky juicy latex when cut which has been traditionally used for the treatment for skin problems, gastrointestinal disorders, respiratory disorders including asthma, bronchitis and hay fever [10,11]. Besides, *E. hirta* is also documented for its biological activities such as antioxidant [12], antibacterial [13], antifungal [14] and anti cancer [12]. Due to its wide usage and availability, the present study was aimed at evaluating *E. hirta* for its phytoconstituents, antibacterial and antioxidant potential.

2. MATERIALS AND METHODS

2.1 Collection of Plant Material

Fresh aerial parts of *E. hirta* were collected from Rohtak district, Haryana. The collected plant material was washed with tap water and then again with distilled water. The plant material was then air dried at room temperature for about 7 days and further dried in oven at 35°C for about 2-3 days. The dried material was grinded with the help of a mixer grinder and powder obtained was stored in air tight containers in refrigerator at 4°C.

2.2 Extraction of Plant Material

The powdered plant material was extracted using Soxhlet apparatus in four different solvents i.e. acetone, water, ethanol and petroleum ether (1:5 W/V) for 2-3 days. The extracts were concentrated by solvent evaporation and each of the extract was weighed and stored at 4°C till further use. Percentage yield of crude extract was calculated as:

Weight of crude extract/weight of powdered plant material X 100

2.3 Test Organisms

Three Gram negative bacterial strains namely *Klebsiella pneumoniae* (MTCC NO 109), *Pseudomonas aeruginosa* (MTCC NO 2453), *Chromobacterium violaceum* (MTCC NO 2656) and three Gram positive bacterial strains namely *Staphylococcus aureus* (MTCC NO 96), *Mycobacterium smegmatis* (MTCC NO 992) and *Bacillus subtilis* (MTCC NO 2057) were procured from IMTECH (Institute of microbial technology), Chandigarh and used in the investigation. Bacterial cultures were inoculated in 10 ml of sterile nutrient agar (NA) medium in culture tubes at 37°C for 24 h. All the cultures were sub cultured monthly on a regular basis and stored at 4°C.

2.4 Disc Diffusion Assay

Disc diffusion assay was carried out as per standard method against all the six bacterial strains used in the present study [15]. Two concentration of each plant extract (200 mg/ml, 100 mg/ml) were prepared by reconstituting the concentrated plant material with the respective solvents. The bacterial strains (1.5×10^8 CFU/ml) were seeded into the respective medium in sterilized Petri Plates. After solidification of medium, sterilized whatman filter paper discs (6 mm in diameter) impregnated with the extract were placed on to the medium separately. Standard disc of ampicillin and blank disc (impregnated with extraction solvents) were also placed on the medium was used as positive and negative control, respectively. Petri Plates were incubated at 37°C for 24 h and the diameter of the inhibition zone obtained was measured. The assay was performed in triplicate for each extract and mean value was recorded.

2.5 Determination of Minimal Inhibitory Concentration (MIC)

MIC was determined by micro broth dilution technique using serially diluted (2 fold) plant extract as per the method of Sarkar et al. [16] with little modifications. 0.1 ml of each plant extract and nutrient broth were added to the wells of a micro titer plate 0.01 ml of standardized inoculums and 0.01 ml of resazurin sodium salt indicator were added into each well and incubated at 37°C for 24 h in a B.O.D incubator. The lowest concentration (highest dilution) of the extract that showed no color change (purple to pink) was regarded as MIC for that particular bacterial strain.

2.6 Antioxidant Activity

The antioxidant activity of the extracts was determined by DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging assay [17]. 2.5 ml of DPPH solution (0.5 mM) was added to 1 ml of different concentrations of plant extract (100 µg/ml to 1000 µg/ml) and incubated in dark for 30 min. After incubation, the absorbance was measured at 517 nm. Ascorbic acid was used as standard. At least three repetitions were run for each assay. The scavenging activity of the extracts was calculated in % inhibition according to the formula given below:

$$\% \text{ Inhibition} = \frac{(\text{A control} - \text{A sample})}{\text{A control}} \times 100$$

2.7 Phytochemical Screening

Qualitative phytochemical screening was carried out with the different plant extracts for the presence of different phytochemicals (phenols, flavonoids, alkaloids, saponins, glycosides, steroids and tannins) as per the standard methods [18].

2.8 Total Phenolic Content (TPC)

Total Phenolic Content was determined as per the method of Aiyegroro and Okoh [19], with some modifications. 1 ml of plant extract was diluted with 5 ml of double distilled water and 1.5 ml of Folin Ciocalteu's reagent was added to it. After 5 min, 4 ml of 20% w/v sodium carbonate solution was added and final volume was made up to 25 ml with double distilled water. Absorbance was recorded at 765 nm after 30 minutes. Gallic acid was used as standard. The amount of phenol in plant extract was calculated by formula given below and expressed as mg/g of equivalent to Gallic acid (mg GAE/g):

$$T = C \times V/M$$

Where,

T = Total Phenolic Content mg/g of Gallic acid equivalent (GAE)

C = Concentration of Gallic acid established from the calibration curve in mg

V = Volume of plant extract in ml

M = Weight of plant extract in gm

2.9 Total Flavonoid Content (TFC)

Total Flavonoid Content was determined by as per the method of Aiyegroro and Okoh [19]. 1 ml

of the extract was taken into a 25 ml volumetric flask and mixed with 1 ml of methanol and 0.1 ml of 10% aluminium chloride. 1 M potassium acetate and 2.8 ml of distilled water was also added. The reaction mixture was allowed to stand at room temperature for 30 minutes. The absorbance was measured at 415 nm. TFC was calculated with the formula given below and was expressed as mg/g of Quercetin equivalent (mg QE/g):

$$T = C \times V/M$$

Where,

T = Total Flavonoid Content mg/g of Quercetin equivalent (QE)

C = Concentration of Quercetin established from the calibration curve in mg

V = Volume of plant extract in ml

M = Weight of plant extract in gm

2.10 Statistical Analysis

Pearson correlation coefficient test was carried out using PAST3 software to analyze the level of correlation between IC 50 values and TPC and between TPC and TFC. Standard deviation was also determined for the zone of inhibition and MIC values of different extracts against selected bacterial strains.

3. RESULTS

In the present investigation, crude extracts prepared from aerial parts of *E. hirta* in four solvents i.e. water, acetone, ethanol and petroleum ether were evaluated for their phytochemical, antibacterial and antioxidant potential.

3.1 Yield of the Crude Extracts

Percentage yield of crude extracts of *E. hirta* obtained was compared for four solvents i.e. acetone, aqueous, ethanolic and petroleum ether extracts. The maximum percentage yield was recorded with water (7.8%) followed by acetone

(5.9%), ethanol (4.6%) and petroleum ether (1.4%) (Table 1).

3.2 Antibacterial Activity

Among four crude extracts of *E. hirta*, three extracts i.e. acetonic, aqueous and ethanolic extracts showed antibacterial activity against all six bacterial strains used in the present study (Table 2). Out of the two concentrations of plant extracts used in the study, i.e. 100 mg/ml and 200 mg/ml, larger zone of inhibition were obtained with 200 mg/ml concentration of plant extract. So, the results obtained with 200 mg/ml concentration are compared among different solvents. Acetone extract showed similar and maximum zone of inhibition against *Bacillus subtilis* and *Chromobacterium violaceum* (17.30±0.57 mm) followed by *Pseudomonas aeruginosa* (15.30±0.57 mm). The extract exhibited similar zone of inhibition against *Klebsiella pneumoniae* and *Mycobacterium smegmatis* (14.30±0.57 mm). Minimum inhibition zone with this extract was observed against *Staphylococcus aureus* (10.60±0.57 mm).

Aqueous extract revealed maximum activity with 14.60±0.57 mm inhibition zone against *Staphylococcus aureus* followed by *Klebsiella pneumoniae* (14.30±0.57 mm) and *Pseudomonas aeruginosa* (14.0±0 mm). The zone of inhibition observed against *Mycobacterium smegmatis* and *Bacillus subtilis* was 11.30±0.57 mm. Least activity was observed against *Chromobacterium violaceum* (7.0±0 mm) with the extract.

Ethanolic extract showed maximum zone of inhibition (17.60±0.57 mm) against *Klebsiella pneumoniae* followed by *Mycobacterium smegmatis* (17.30±0.57 mm) and *Pseudomonas aeruginosa* (16.60±0.57 mm). The extract showed similar zone of inhibition against two bacterial strains i.e. *Chromobacterium violaceum* and *Staphylococcus aureus* (12.60±0.57 mm). The crude extract prepared in petroleum ether was found to be effective only against *Chromobacterium violaceum* (12.60±0.57 mm).

Table 1. Percentage yield of the crude extracts of *E. hirta*

Solvents	Raw plant powder (gm)	Crude extract (gm)	Percentage yield (%)
Acetone	100	5.9	5.9
Aqueous	100	7.8	7.8
Ethanol	100	4.6	4.6
Petroleum ether	100	1.4	1.4

Table 2. Zone of inhibition obtained against six bacterial strains with crude extracts of *E. hirta* in different solvents

Bacterial strains used	Zone of inhibition diameter (mm)								
	Acetone extract (mg/ml)		Aqueous extract (mg/ml)		Ethanollic extract (mg/ml)		Petroleum ether extract (mg/ml)		Positive control (Ampicillin) (µg/ml)
	100	200	100	200	100	200	100	200	100
<i>Klebsiella pneumoniae</i>	12.30±0.57	14.30±0.57	12.30±0.57	14.30±0.57	15.30±0.57	17.60±0.57	-	-	24.33±2.51
<i>Pseudomonas aeruginosa</i>	14.30±0.57	15.30±0.57	10.60±0.57	14.0±0	15.30±0.57	16.60±0.57	-	-	22.0±4.0
<i>Staphylococcus aureus</i>	8.60±0.57	10.60±0.57	13.30±0.57	14.60±0.57	9.30±0.57	12.60±0.57	-	-	15.33±1.52
<i>Mycobacterium smegmatis</i>	10.0±0	14.30±0.57	9.60±0.57	11.30±0.57	15.30±0.57	17.30±0.57	-	-	28.33±1.52
<i>Bacillus subtilis</i>	13.60±0.57	17.30±0.57	10.0±0	11.30±0.57	11.30±0.57	12.30±0.57	-	-	24.33±1.52
<i>Chromobacterium violaceum</i>	14.60±0.57	17.30±0.57	7.0±0	7.0±0	12.60±0.57	12.60±0.57	12.30±0.57	12.60±0.57	22.33±1.52

Values are mean ± SD of the three replicates

MIC was performed only with those plant extracts that exhibited antibacterial activity with disc diffusion assay. MIC value for different crude extracts of *E. hirta* ranged between 0.12±0.04 mg to 5.0±0 mg (Table 3). Least MIC value was obtained against *Klebsiella pneumoniae* with ethanolic extract (0.12±0.04 mg) followed by acetone extract and aqueous extract (0.62±0 mg). For *Pseudomonas aeruginosa*, minimum MIC value was observed with ethanol extract (0.41±0.17 mg) followed by acetone extract (0.51±0.17 mg) and aqueous extract (1.25±0 mg). Aqueous extract exhibited least MIC value (0.51±0.17 mg) for *Staphylococcus aureus* followed by ethanol extract (0.62±0 mg) and acetone extract (1.25±0 mg). Ethanol extract showed minimum MIC value against *Mycobacterium smegmatis* (0.20±0.09 mg) followed by acetone extract (0.83±0.36 mg) and aqueous extract (2.50±0 mg). MIC values obtained with acetone extract, ethanolic extract and aqueous extract against *Bacillus subtilis* were 0.20±0.09, 1.25±0 and 2.50±0 mg, respectively. Acetone extract showed low MIC value (0.20±0.09 mg) with *Chromobacterium violaceum* as compared to ethanol extract (0.62±0 mg) and petroleum ether extract (1.25±0 mg).

3.3 Antioxidant Activity

When the crude plant extracts of *E. hirta* were investigated for their free radical scavenging

activity, aqueous extract exhibited maximum free radical scavenging activity followed by acetone extract, ethanol extract and petroleum ether extract (Fig. 1). Similarly, minimum IC 50 value (105.39 µg/ml) was obtained with aqueous extract followed by acetone extract (371.0 µg/ml), ethanol extract (504.92 µg/ml) and petroleum ether extract (645.12 µg/ml) (Fig. 2).

3.4 Phytochemical Screening

The results obtained with four crude extracts of *E. hirta* prepared in four different solvents are given in Table 4.

3.5 Total Phenolic Content

Total Phenolic Content is reported as Gallic acid equivalents (GAE) by reference to standard curve ($y=0.0006x$, $r^2 =.9926$) (Fig. 3). Out of the four extracts, aqueous extract showed highest TPC value i.e. 8.2 mgGAE/g, followed by acetone extract (6.4 mgGAE/g) and ethanol extracts (5.84 mgGAE/g). Lowest TPC value was exhibited by petroleum ether extract (2.23 mgGAE/g) (Fig. 4).

3.6 Total Flavonoid Content

Total Flavonoid Content was determined as Quercetin equivalents (QE) by reference to standard curve ($y=0.0006x$, $r^2 =.9874$) (Fig. 5).

Table 3. MIC values obtained against six bacterial strains with crude extracts of *E. hirta* prepared in different solvents

Bacterial strains	Solvent	MIC value (mg)
<i>Klebsiella pneumoniae</i>	Acetone	0.62±0
	Aqueous	0.62±0
	Ethanol	0.12±0.04
<i>Pseudomonas aeruginosa</i>	Acetone	0.51±0.17
	Aqueous	1.25±0
	Ethanol	0.41±0.17
<i>Staphylococcus aureus</i>	Acetone	1.25±0
	Aqueous	0.51±0.17
	Ethanol	0.62±0
<i>Mycobacterium smegmatis</i>	Acetone	0.83±0.36
	Aqueous	2.50±0
	Ethanol	0.20±0.09
<i>Bacillus subtilis</i>	Acetone	0.20±0.09
	Aqueous	2.50±0
	Ethanol	1.25±0
<i>Chromobacterium violaceum</i>	Acetone	0.20±0.09
	Aqueous	5.0±0
	Ethanol	0.62±0
	Petroleum ether	1.25±0

Values are mean ± SD of the three replicates

Aqueous extract showed highest TFC i.e. 4.6 (2.05 mgQE/g), ethanol (1.82 mgQE/g) and mgQE/g, followed extracts prepared in acetone Petroleum ether i.e. 0.79 mgQE/g (Fig. 6).

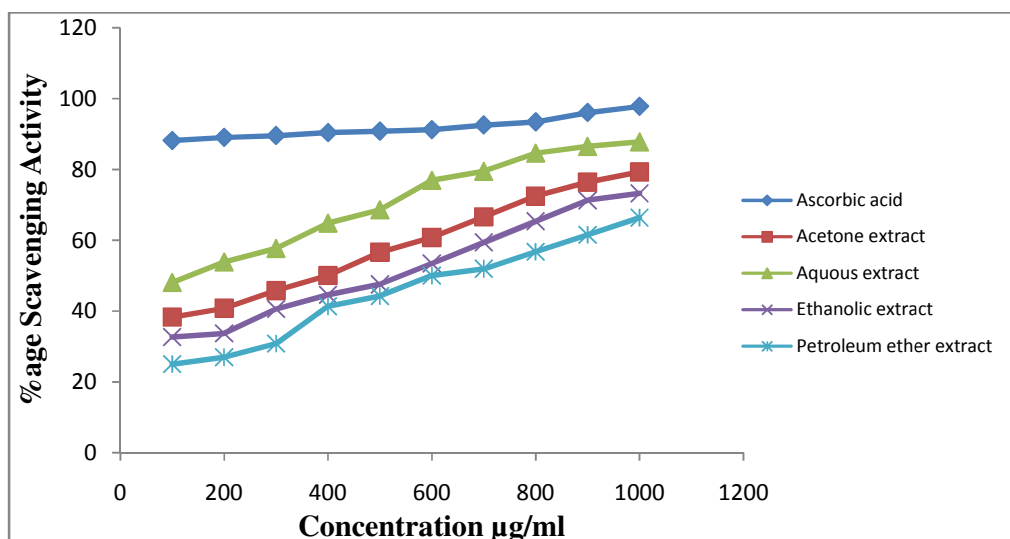


Fig. 1. % age scavenging activity obtained against different concentrations of *E. hirta* plant extracts and ascorbic acid

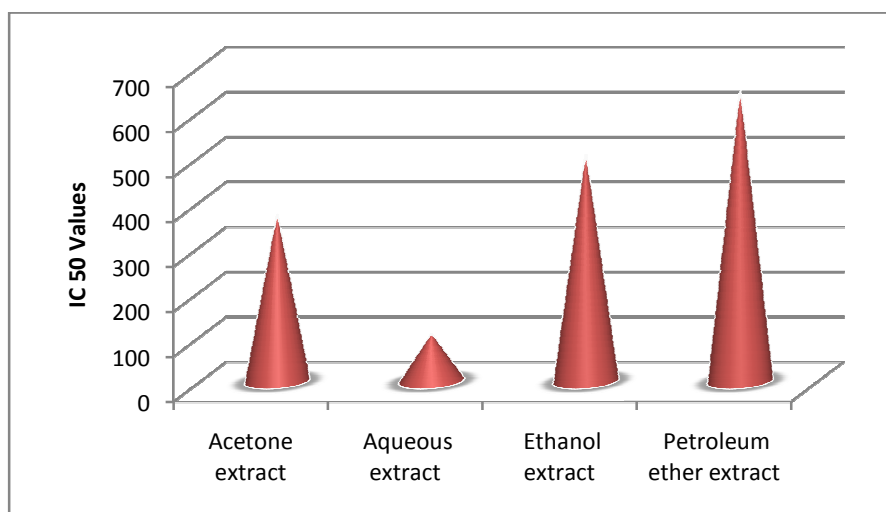


Fig. 2. IC 50 values obtained with different extracts of *E. hirta*

Table 4. Phytochemical analysis of different extracts of *E. hirta*

Phytochemicals tested	Plant extracts			
	Acetone	Aqueous	Ethanol	Petroleum ether
Alkaloids	++	+	++	+
Phenols	+++	+++	+++	+
Flavonoids	++	++	++	+
Glycosides	+	+	+	-
Tannins	++	+	++	+
Saponins	+	+	-	-
Steroids	++	+	+	-

Presence: +, Absence: -

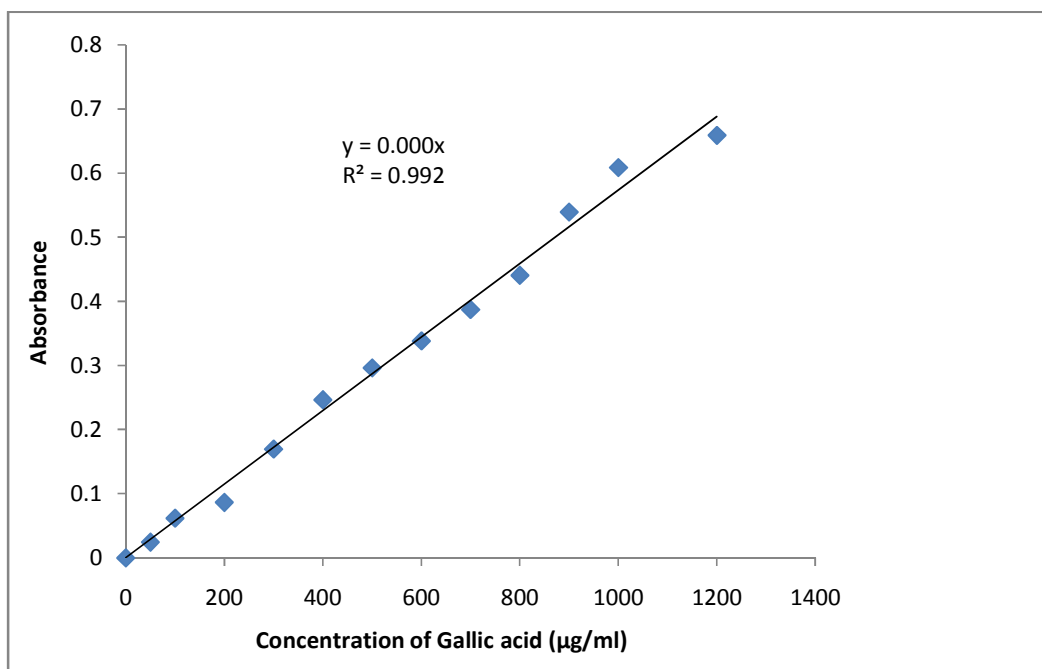


Fig. 3. Gallic acid standard curve

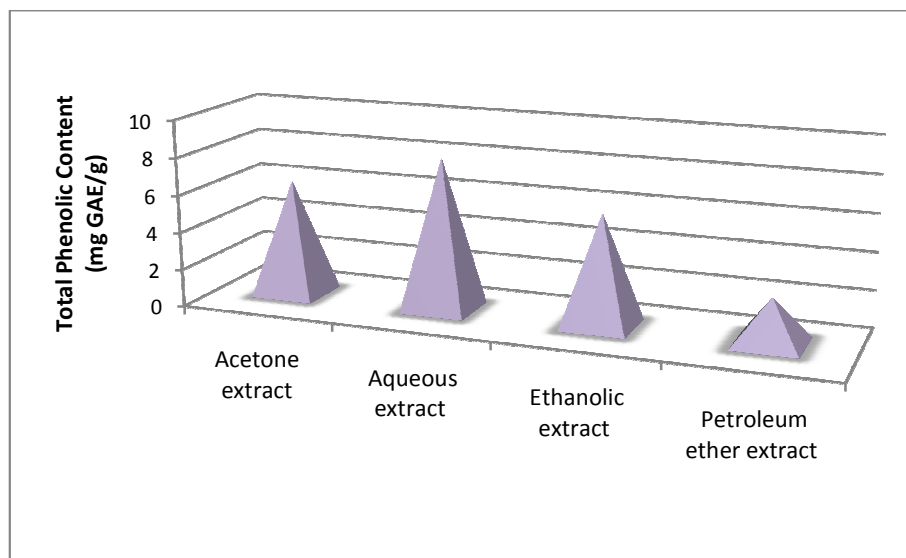


Fig. 4. Total phenolic content obtained for different extracts of *E. hirta*

Correlation coefficient between IC 50 values and total phenolic content of different extracts was found to be -0.918 which shows these two variables to be negatively co-related. Correlation coefficient between total phenolic content and total flavonoids content (0.883) showed that these two variables are positively co-related.

4. DISCUSSION

E. hirta holds a very significant position in traditional system of medicine due to its extensive capability to alleviate a number of health disorders. The present investigation was therefore planned to evaluate four crude extracts of *E. hirta* for their antibacterial, antioxidant and phytochemical potential.

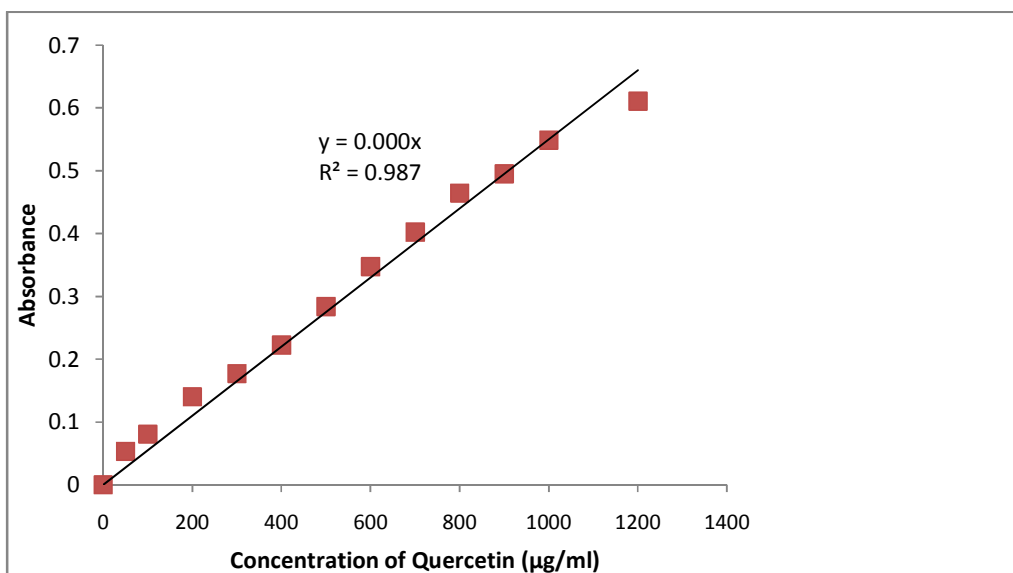


Fig. 5. Quercetin standard curve

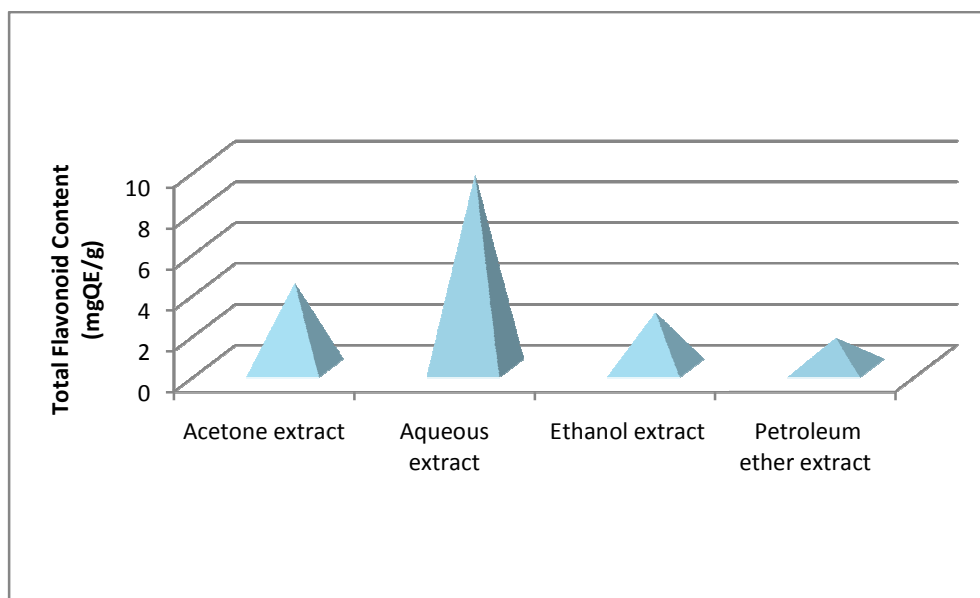


Fig. 6. Total flavonoid content obtained for different crude extracts of *E. hirta*

Out of the four extracts, three extracts (in acetone, water and ethanol) were found to be effective against both Gram positive and Gram negative bacterial strains, though to different degrees thus showing variable susceptibility of microorganisms. Similar to our findings, other workers have also reported acetonic plant extract of *Euphorbia hirta* to be effective against both Gram negative (*K. pneumoniae*) and Gram positive bacteria (*S. aureus* and *B. subtilis*) [20]. Masih et al. [21] also reported significant

antibacterial activity of acetonic and ethanolic extract of *E. hirta* against Gram negative (*Escherichia coli*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*) and Gram positive bacteria (*Staphylococcus aureus*). *E. hirta* has also been shown to possess strong antibacterial activity than other species of *Euphorbia* and this was attributed to the presence of tannin, alkaloids and flavonoids [22]. Maximum inhibition zone and accordingly minimum MIC value with acetone extract was observed against *B. subtilis*

and *C. violaceum* followed by *P. aeruginosa*, *K. pneumoniae*, *M. smegmatis* and *S. aureus*.

For aqueous extract, maximum diameter of inhibition zone was obtained against *S. aureus* and minimum MIC value was also observed against it. The crude extract also exhibited significant inhibition zone and MIC values against *K. Pneumoniae* and *P. aeruginosa*. Interestingly, similar inhibition zone and MIC value were obtained against *B. subtilis* and *M. smegmatis* but they were lower than the above mentioned bacteria. The plant extract was found to be least effective against *C. violaceum*. Similar to our results, other workers also reported significant activity of aqueous extract against different bacterial strains. Patel and Patel [23] reported significant zone of inhibition against *P. aeruginosa*. Abubakar [24] also observed better antibacterial activity of aqueous extracts as compared to the extracts in the organic solvents. Both aqueous and ethanolic leaf extracts of *Euphorbia hirta* have been reported to be effective against *K. pneumoniae*, *P. aeruginosa*, *S. aureus* and also against other bacterial strains like *Escherichia coli*, *Proteus vulgaris*, *Salmonella typhi* and *Shigella dysenteriae* [25].

Least MIC value and maximum zone of inhibition was observed with ethanolic extract against *K. pneumoniae* followed by *M. smegmatis* and *P. aeruginosa*. The extract showed similar antibacterial activity against *S. aureus* and *C. violaceum*. Similar to our observations, antibacterial activity of ethanolic extract of *E. hirta* has been reported against *Bacillus pumilus*, *S. aureus*, *Streptococcus pneumoniae*, *E. coli* and *K. pneumoniae* [22].

Out of the four extracts, petroleum ether extract possessed poor antimicrobial activity as compared to other extracts. This is probably because of the low percentage yield of crude extract in petroleum ether. However, contrary to our results, Patel and Patel [23] reported good antibacterial activity of petroleum ether extract against many bacterial strains including *K. pneumoniae*. Although it difficult to explain this, but according to us this could be due to factors like collection of plant material from different geographical area, age of the plant and variations in the processing of the material.

Ampicillin used as positive control produced larger zone of inhibition even at a very low concentrations as compared to crude extracts. The main reason appears to be the fact that

synthetic antibiotics are pure chemical compounds whereas the crude extract contains some impurities which may be inert and devoid of antibacterial activities [24]. MIC values of different solvents against 6 bacterial strains were corresponding to their respective zone of inhibition observed by disc diffusion assay.

Plant synthesizes an array of secondary metabolites such as phenols, tannins, terpenoids, alkaloids and flavonoids which are of great benefit to mankind due to their therapeutic potential. Therefore, medicinal properties of plants vary from species to species as it depends upon the presence and absence of these secondary metabolites [26]. Phytochemical screening of crude extracts of *E. hirta* revealed the presence of bioactive compounds like phenols, flavonoids, alkaloids, saponins, glycosides, steroids and tannins known for their medicinal and curative properties. Phenolics and flavonoids are responsible for the antioxidant activity of the plant. Different workers have also reported the presence of these phytoconstituents from plant extracts of *E. hirta* [20,24,27-30].

When different extracts were evaluated for their antioxidant activity by DPPH radical scavenging method, aqueous extract was found to exhibit highest percentage scavenging activity and lowest IC 50 value. The total phenolic and flavonoid content of the aqueous extract was also significantly high and it shows that water is a good solvent for the extraction of bioactive compounds. Similar to our results, other workers have also reported high antioxidant activity of aqueous extract as compared to organic solvents. In fact, antioxidant activities of aqueous extract of *E. hirta* are found to be comparable to that of green and black tea [31]. Other workers also reported significant scavenging activity of aqueous extract [32,33]. Out of the three extracts prepared in organic solvents, acetonic extract showed maximum activity which again corresponds to its high phenolic content. Mishra et al. [34] also reported ethanolic plant extract to exhibit good antioxidant activity and a direct relation of it with phenolic content. Ethanolic and petroleum ether extract of even flowers of *E. hirta* have been reported to possess good antioxidant activity [35].

5. CONCLUSION

The present in vitro study revealed significant antibacterial and antioxidant activity of crude extract of *E. hirta* in acetone, water and ethanol.

Total phenolic and flavonoid content of different extracts exhibited a significant positive correlation with scavenging activity of the respective extracts. As the crude extracts of *E. hirta* possess good antibacterial and antioxidant activity, hence it may be helpful in the treatment of various disorders due to microbes and free radicals. However, further phytochemical and pharmacological studies are needed to isolate and characterize the bioactive constituents of the plant.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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