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ANTIBACTERIAL POTENTIAL AND PHYTOCHEMICAL INVESTIGATION OF MEDICINAL PLANT (*EUPHORBIA HIRTA LINN)*

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ABSTRACT

Euphorbia hirta linn (*E. hirta*) is a medicinal plant, also known as asthma weeds which are mainly used against human pathogenic diseases such as digestive tract problem, asthma, coughs, diarrhea, dysentery, typhoid fever, inflammation, chronic bronchitis, and other pulmonary disorders. The native of these plants is in Asia, Australia, and East and West Africa. In the present study antibacterial activities, antioxidant activities, and phytochemicals were estimated from 20% isopropanol and aqueous extract of different parts (leaf, fruit, stem, root) of *E. hirta*. The maximum antioxidant activity, phenolic contents, and flavonoids showed 0.418 mg/ml in aqueous fruit extract, 0.176 and 0.43 mg/ml in aqueous leaf extract respectively. Some other bioactive compounds were also observed from both 20% extracts of different segments of *E. hirta* qualitatively. Furthermore, some biomolecules were estimated in higher amounts from *E. hirta* plant extracts as total sugar 0.168 mg/ml from the aqueous extract of the fruit, total protein 0.453 mg/ml from isopropanol extract of the fruit, and the reducing sugar contents 0.08 mg/ml from aqueous root extract. Different parts of the understudy plant (*E. hirta*) extracts exhibited antibacterial activity against *E. coli and K. pneumonia*. Whereas only aqueous and isopropanol extract of root and stem and aqueous extract of fruit showed negative antibacterial activity against *S. pneumonia*. The result suggested that *E. hirta* may be used for the treatment of different infectious diseases due to the presence of different phytochemicals.

Key Words: *Euphorbia hirta, antibacterial activity, antioxidants, phytochemicals*

INTRODUCTION

Plants always have extraordinary benefits for society. Plants are used by human beings for their basic requirements like feeding, clothing, sheltering, hunting, and nursing (Vitalini, Tomè, & Fico, 2009). A complete store of medicines has been gifted in the form of plants by nature to cure all diseases of mankind (Jana, Rahaman, & Banerjee, 2009). Medicinal plants have a pivotal role in human health (H. Soni, Sharma, & Malik, 2020). According to WHO (Organization, 1991), medicinal plant is considered those plant that contains ingredients that are used for ailment-curing purposes. According to World Health Organization, in developing countries, about 80% of the population still depends on traditional plant-based medicines for the prevention and curing of diseases (Ikegami, Fujii, Ishihara, & Satoh, 2003). Various parts (leaves, roots, seeds etc.) of medicinal plants extracts have long been used since ancient times for treating various human pathogenic diseases (Aqib, 2021). It is documented that four out of five persons depend on traditional medicine for their basic healthcare necessities worldwide (Kebede, Gadisa, & Tufa, 2021). Although in the past, various antimicrobial compounds were discovered from natural and synthetic products for the control and treatment of pathogens (Shriram, Khare, Bhagwat, Shukla, & Kumar, 2018), only some of them were available in world market (Poulakou, Lagou, Karageorgopoulos, & Dimopoulos, 2018). Many studies have revealed that medicinal plants performed several biological activities due to presence of secondary metabolites include several groups of molecules, including alkaloids, lignans, steroids, phenolic, glycosides and carbohydrates etc. (Tran, Nguyen, *et al.*, 2020). These bioactive compounds are used as initial material for synthesis of antibiotics to treat infectious diseases (Rahman & Anwar, 2007). In the traditional system, plant based medicines are extensively used for the treatment of gastrointestinal ailments, respiratory diseases, wound healing, tumors, lactation in women and urogenital

ailments(K. R. Sharma, 2020). The herbal medicine or therapeutic agents originated from plant and are known as important and alternate source of conventional medicines for treatment of various diseases (Jachak & Saklani, 2007; Sasidharan, Chen, Saravanan, Sundram, & Latha, 2011). However, till now less than 1% plants' phytochemicals recognized as secondary metabolites and pharmacologically active component (Motaleb, 2011). In this connection, traditional medicinal plants are the utmost treasured source of new bioactive chemical articles due to their chemical ecological biodiversity and different chemical constituents (Kenneth-Obosi & Babayemi, 2017). Singh *et al*., (2021) have investigated the phytochemicals obtained from plant sources have been utilized for the prohibition and cure of various ailments including diarrhea, heartburn, peptic ulcers, vomiting, bronchitis, asthma, kidney stones coughs, colds, sterility, menstrual problems and sexually transmitted diseases. According to WHO, approximately 21,000 plant species are considered as medicinal plants, however more than 30% of the whole plant species are already in practice (Khan, Khan, Ullah, & Nadhman, 2021). *Euphorbia hirta linn* (*E. hirta)* is a small annual herb, found in humid regions, belongs to Euphorbiaceae family (Jachak & Saklani, 2007). It is frequently recognized as milkweed and an asthma plant. In different countries of the world. The plant contains milky white latex, which has variable toxicity. *E. hirta* displays different pharmacological actions due to the presence of flavonoids, polyphenols, alkaloids, triterpenoids and saponins. Conventionally, the plant is used for the treatment of vomiting, peptic ulcers, dysentery, diarrhea, scabies, menstrual problems, tinea, thrush, measles, fungal diseases and as an antibacterial to treat sores, wounds and conjunctivitis (Singh, Chaubey, & Mishra, 2021). *E. hirta* is famous as an analgesic to treat rheumatism, toothache, severe headache, colic and pains during pregnancy and as an antidote and pain relief of snakebites and scorpion stings. Furthermore, in the treatment of diabetes, this plant was used as a folk medicine for a long time (Sheliya *et al.*, 2016; Tran, Tran, Truong, & Le, 2020). In addition, recent pharmacological research revealed that *E. hirta* possess several pharmacological properties such as antidiabetic (Subramanian, Bhuvaneshwari, & Prasath, 2011), anticancer, antioxidant, and anti-inflammatory (N. Sharma *et al.*, 2014), anthelmintic, antidipsogenic, angiotensin-converting enzyme inhibitory, anti-arthritis, and galactogenic (Al-Snafi, 2017), wound healing potential (Tuhin *et al.*, 2017), hepatoprotective (Tiwari, Mishra, Bhatt, & Chaudhary, 2015), antibacterial (Kader, Noor, Radzi, & Wahab, 2013), antianaphylactic (Youssouf *et al.*, 2007) and anxiolytic (Xia *et al.*, 2018). Moreover, all over the world, herbs are considered more effective, economical, safe, reliable and easily available natural resources of drugs. So the main purpose of this research work is to assess the qualitative and quantitative screening of phytochemicals such as flavonoid and phenolic, the potential of *E. hirta* against some selected Gram positive and negative bacterial species and investigated the in vitro antioxidant activity and evaluate its potential for clinical use as a natural antioxidant.

MATERIALS AND METHODS

Collection of Plants and 20% Aqueous and Isopropanol Extract: *E. hirta* plant was collected from surrounding of Jamshoro. The collected plant *E. hirta* was washed with tape water. Different parts of plant (root, stem, leaf and fruit) were separated and dried. From dried samples of root, stem, leaf and fruit, 20% aqueous and isopropanol extracts were prepared by reported method (Dahot, 1999). The pH of extracts was determined by pH meter and finally stored at 4ºC.

Gram Staining Method and Preparation of Inoculum: The Gram positive *Staphylococcus aureus* (*S. aureus)*, Gram negative bacteria *Escherichia coli* (*E. coli*) and *Klebsiella pneumonia* (*K. pneumonia*)were pre-cultured in Luria Bertani medium for overnight at 37°C.The smear of species on the glass slide was prepared by Gram staining method (Gram, 1884).

Antibacterial Activity Analysis: Antibacterial activity was determined from prepared extracts by agar well diffusion method (Mothana & Lindequist, 2005). The control was calculated by using antibiotics (Piperacillin/Tazobactam 110 µg, Avelox 5 µg, Oxacillin 1µg, Erythromycin 15µg, and Ofloxacin 5 µg). Antibacterial activity was tested against *E. coli*, *S. aureus* and *K. pneumonia* by well formation method (Naqvi *et al.*, 2011).

Quantification Of Antioxidant Activity: Antioxidant activity of *E. hirta* plant was quantified by the reported method (Prieto, Pineda, & Aguilar, 1999). 0.2ml of each sample of both water and isopropanol extracts of roots, stem, leaf and fruit of *E. hirta* combined with 2 ml of reagent solution (0.6 M sulphuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). The samples tubes were capped with aluminum foil and incubated in the boiling water bath at 95°C for 90 minutes. The samples were cooled at room temperature and the absorbance was read at 695 nm against the blank.

Quantification of Phenolic and Flavonoid Content: The amount of total phenolic was quantified by the Folin-ciocalteu method (Yasoubi, Barzegar, SAHARI, & Azizi, 2007). The absorbance of samples was read against the blank at 765 nm and the results were calculated from the Gallic Acid standard curve. The total flavonoid content (TFC) was calculated according to the reported aluminum chloride method (Djeridane *et al.*, 2006). The absorbance was monitored against blank at

510 nm. The TFC was estimated from the standard calibration curve of Quercetin.

Quantification of Total Sugar, Reducing Sugar and Total Protein: The total sugar from 20% isopropanol and aqueous extracts of leaf, fruit, stem and root of *E. hirta* plant was determined (Montogomery, 1961). The absorbance was read against blank by UV-visible spectrophotometer at 485 nm. The glucose standard curve was used for calculation of total sugar concentration from test sample. The reducing sugar estimated by reported Dinitrosalicylic acid (DNS) method (Miller, 1959). The color intensity was read against blank by UV-visible spectrophotometer at 540nm. Blank was prepared by adding distilled water in substitution of test sample. The total protein from 20% isopropanol and aqueous extracts of leaf, fruit, stem and root of *E. hirta* plant was quantified according to protocol described by Lowery et al., (Reagent, 1951). Absorbance of each sample was read against blank at 750 nm. The results were prepared by albumin standard curve.

Qualitative Test of Phytochemicals: Qualitative test of phytochemicals were also determined by reported methods such as alkaloids, tannins, coumerin, steroids (A. Soni & Sosa, 2013), flavonoids (Njoku & Obi, 2009), cardiac glycosides (C. glycosides) (Yadav & Agarwala, 2011) and terpenoids (Edeoga, Okwu, & Mbaebie, 2005) from different parts (root, stem, fruit and leaf) of *E. hirta*.

RESULTS

The plant extracts and phytochemicals are utilized as antimicrobial properties that have important roles for treatment of diseases. Many researches have been led in different countries to evidence plants efficiency from the previous years. Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the secondary metabolism of the plant (Nascimento, Locatelli, Freitas, & Silva, 2000). Antioxidant properties found in medicinal plants which leads to antibacterial activities. The pH of different parts of *E. hirta* was checked from both 20% isopropanol and aqueous extracts. It was noted that different parts of plants extracts showed pH near to neutral or slightly acidic in nature. The pH was recorded 7.3, 6.3, 5.8 and 5.9 in aqueous root, stem, fruit and leaf extracts from *E. hirta* respectively and 6.1, 5.8, 5.4 and 5.1 in isopropanol root, stem, fruit and leaf extract respectively.

In present study antibacterial activities of different extracts were evaluated by the agar well diffusion method (Mothana & Lindequist, 2005). 20% solvent extracts of different parts of *E. hirta* (root, stem, leaf and fruit) possess good antibacterial activity against selected bacterial species *S. aureus*, *E. coli* and *K. pneumonia*, compared with standard antibiotics (some available in market) as presented in Table-1. The potential of different extracts against the bacterial species was determined in terms of zone inhibition of bacterial growth by scale in millimeter unit.

The results showed the wide difference in the antibacterial activities of *E. hirta* from aqueous and isopropanol extracts. According to the results from agar well diffusion methods, the negative results was showed against *S. aureus* in aqueous root, stem, fruit and in isopropanol root and stem extracts of *E. hirta*, while in aqueous leaf, isopropanol leaf and fruit extracts showed (18 mm, 13.2 mm and 10 mm) respectively. Aqueous and isopropanol root, stem, leaf and fruit extract of *E. hirta* exhibited antibacterial activities against *E. coli* (11.3 mm and 17.3 mm), (12.3 mm and 25.3 mm), (15.3 mm and 16 mm) and (11.3 mm and 16 mm) respectively. The zone of inhibition was showed towards *K. pneumonia* in plant extracts that is revealed in isopropanol root, leaf and fruit extracts of *E. hirta* (10.75 mm, 8 mm and 9.5 mm) and in aqueous leaf and fruit 9.5 mm, 6 mm in that order. The standard results were reported for antibiotics against three species of bacteria. Piperacillin/Tazobactam showed –ve, 5 mm and 5 mm towards *S. aureus, E. coli* and *K. pneumonia*. Avelox revealed 12 mm, -ve and 20.5 mm against *S. aureus, E. coli* and *K. pneumonia*. Oxacillin displayed –ve result towards *S. aureus, E. coli* and 5 mm for *K. pneumonia*. Erythromycin presented highest antibacterial activity for all bacteria 15, 18 and 20 mm respectively. Ofloxacin showed 14.8 mm against *S. aureus* and negative for *E. coli.*

Table- 1: Antibacterial activity of *E. hirta* solvent extracts

Plant extracts	***S. aureus***	***E. coli***	***K. pneumonia***
Isopropanol root extracts	Negative	17.3 mm	10.75 mm
Isopropanol stem extracts	Negative	25.3 mm	10 mm
Isopropanol leaf extracts	13.2 mm	16 mm	8 mm
Isopropanol fruit extracts	10 mm	16.3 mm	9.5 mm
Aqueous root extracts	Negative	11.3 mm	9.5 mm
Aqueous stem extracts	Negative	12.3 mm	9.5 mm
Aqueous leaf extract	18 mm	15.3 mm	9.5 mm
Aqueous fruit extracts	Negative	11.3 mm	6 mm

The different parts extract of *E. hirta* were evaluated qualitatively for the phytochemical constituents which revealed the presence of alkaloids, flavonoids, tannins, coumarin, terpenoids, steroids and C. glycoside by the indication of different colours (Table-2).

Table -2: Qualitative analysis of phytochemicals from *E. hirta* solvent extracts

Plant Extracts	Alkaloid	Flavonoid	Tannins	Coumarin	Terpenoid	Steroid	C.glycosids
Isopropanol root extracts	++	++	++	+	++	++	++
Isopropanol stem extracts	++	++	++	++	++	++	++
Isopropanol leaf extracts	++	+++	+++	++	+++	++	++
Isopropanol fruit extracts	++	+++	+++	++	+++	+++	++
Aqueous root extracts	+	-	++	++	+	+	++
Aqueous stem extracts	+++	+++	+ ++	+ ++	++	++	++
Aqueous leaf extract	+++	+++	+++	++	+	+++	+
Aqueous fruit extracts	++	++	++	+++	+++	+++	+++

Maximum (+++), Moderate (++), Minimum (+), Negative (-)

The results of antioxidant activity from *E. hirta* extracts are shown in Fig.1. The antioxidant contents 0.222 mg/ml, 0.40 mg/ml, 0.391 mg/ml and 0.392 mg/ml were found in isopropanol extracts of root, stem, leaf and fruit respectively whereas in the aqueous extracts of root, stem, leaf and fruit were 0.26 mg/ml, 0.39 mg/ml, 0.318 mg/ml and 0.418 mg/ml respectively.

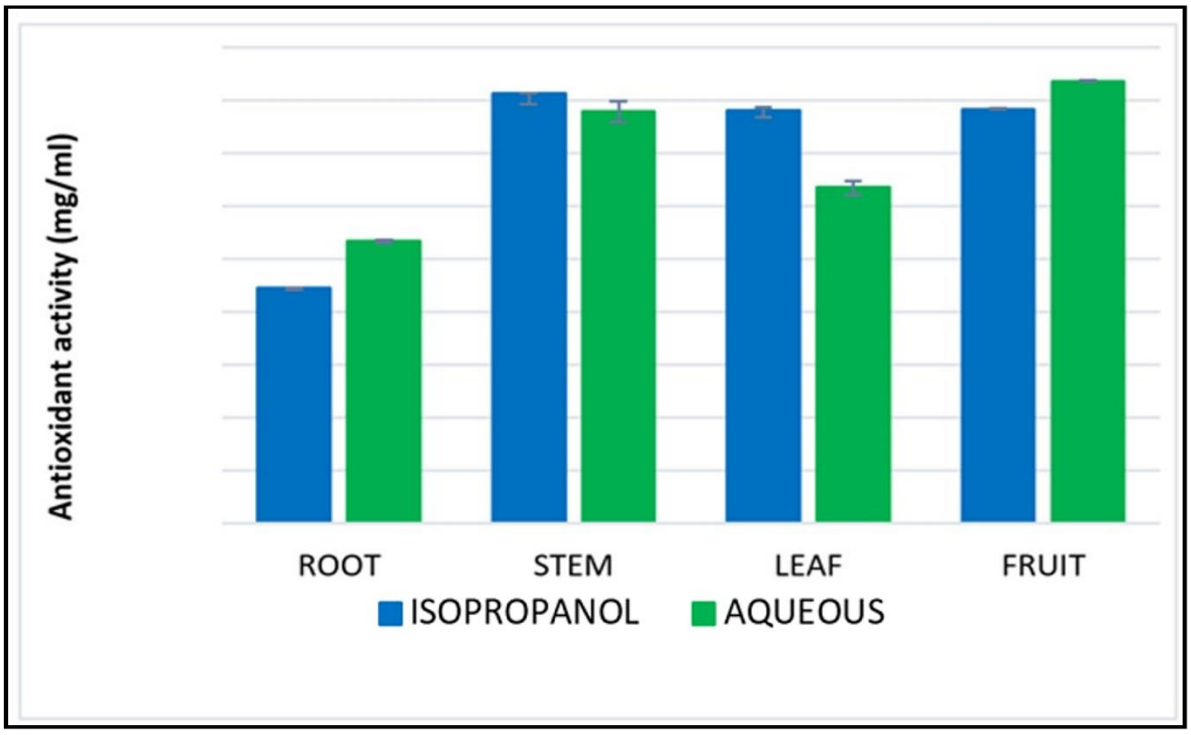

Figure.1 Quantification of antioxidant activity from extracts of *E. hirta*

The phenolic contents in isopropanol extracts of root, stem, leaf and fruit were estimated to be 0.157 mg/ml, 0.163 mg/ml, 0.166 mg/ml and 0.153 mg/ml respectively whereas in the aqueous extracts of root, stem, leaf and fruit they were 0.086 mg/ml, 0.151 mg/ml, 0.176 mg/ml and 0.165 mg/ml respectively as exhibited in Fig.2.

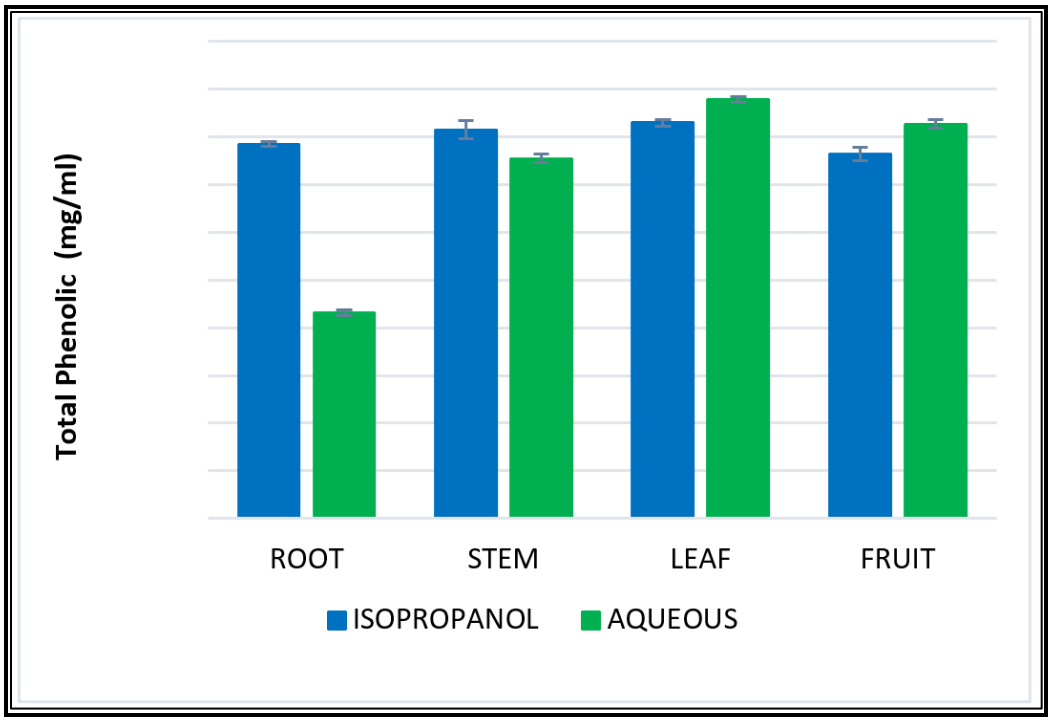

Figure 2. Quantification of phenolic compounds from extracts of *E. hirta*

The flavonoids contents were revealed 0.17 mg/ml, 0.21 mg/ml, 0.35 mg/ml and 0.28 mg/ml in isopropanol extracts of root, stem, leaf and fruit correspondingly, whereas in the aqueous extracts of root, stem, leaf and fruit they were investigated 0.24 mg/ml, 0.26 mg/ml, 0.43 mg/ml and 0.33 mg/ml respectively. Overall it could be concluded that aqueous extracts revealed highest flavonoid contents as compared to isopropanol extract (Fig. 3).

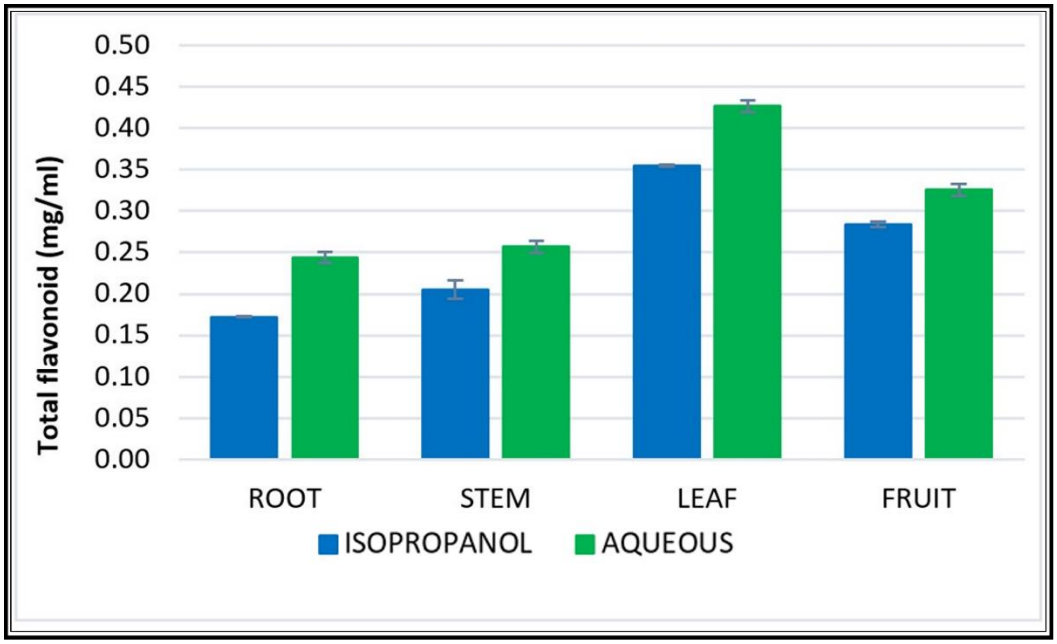

Figure 3. Quantification of total flavonoids from extracts of *E. hirta*

The total sugar contents of different parts of plant extracts were investigated. The maximum concentration of total sugar was observed 0.168 mg/ml and 0.157 mg/ml from aqueous and isopropanol extracts of fruit

and also isopropanol extract of stem of *E. hirta* respectively. Whilst lowest concentration of total sugar 0.128 and 0.130 mg/ml was recorded in both extracts of *E. hirta* leaf (Fig. 4).

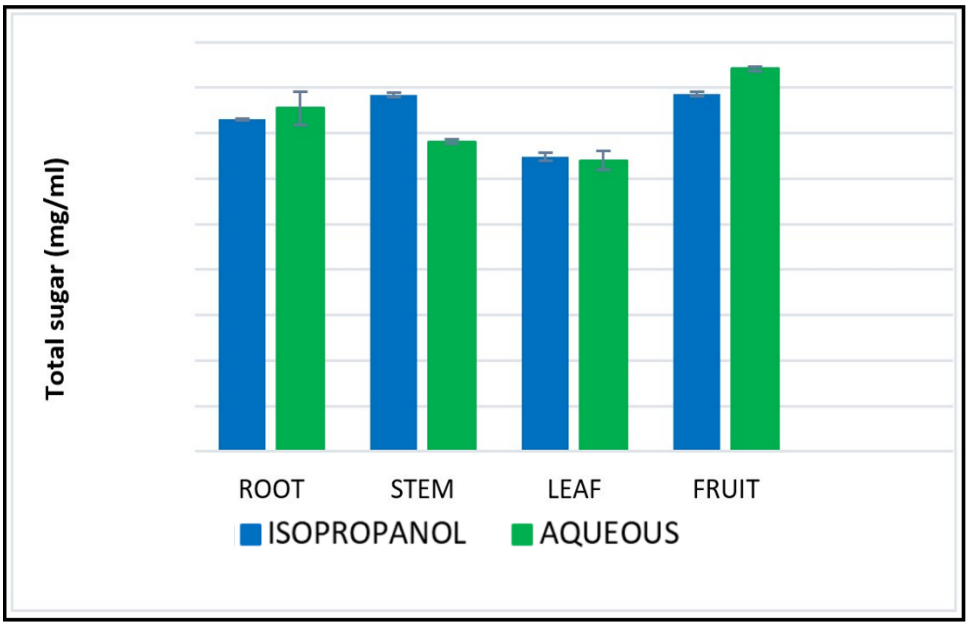

Figure.4 Quantification of total sugar from extracts of *E. hirta*

The reducing sugar contents were noted maximum 0.08 mg/ml in 20% aqueous root extract of *E. hirta* in comparison to other parts of same *E. hirta* plant as reported in Fig. 5. However, the other parts of *E. hirta* extracts showed quite least amount of reducing sugar as aqueous extract of leaf and fruits 0.049 mg/ml, while isopropanol roots and leaf extract 0.046 and 0.048 mg/ml respectively.

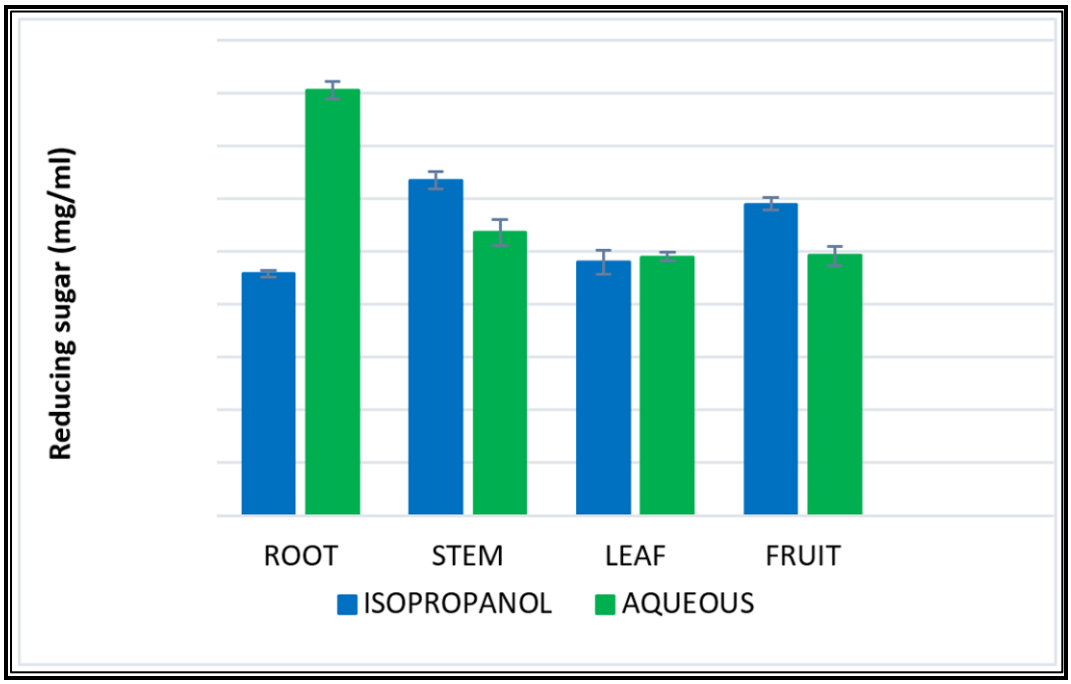

Figure.5 Quantification of reducing sugar from extracts of *E. hirta*

Total protein contents are shown in Fig.6. The protein contents 0.133 mg/ml, 0.430 mg/ml, 0.132 mg/ml and 0.453 mg/ml were observed in isopropanol extracts of root, stem, leaf and fruit respectively, whereas the aqueous extracts of root, stem, leaf and fruit they were 0.118 mg/ml, 0.337 mg/ml, 0.170 mg/ml and 0.443 mg/ml respectively. Overall it was observed that total protein was found to be highest in all isopropanol extracts than in aqueous extracts.

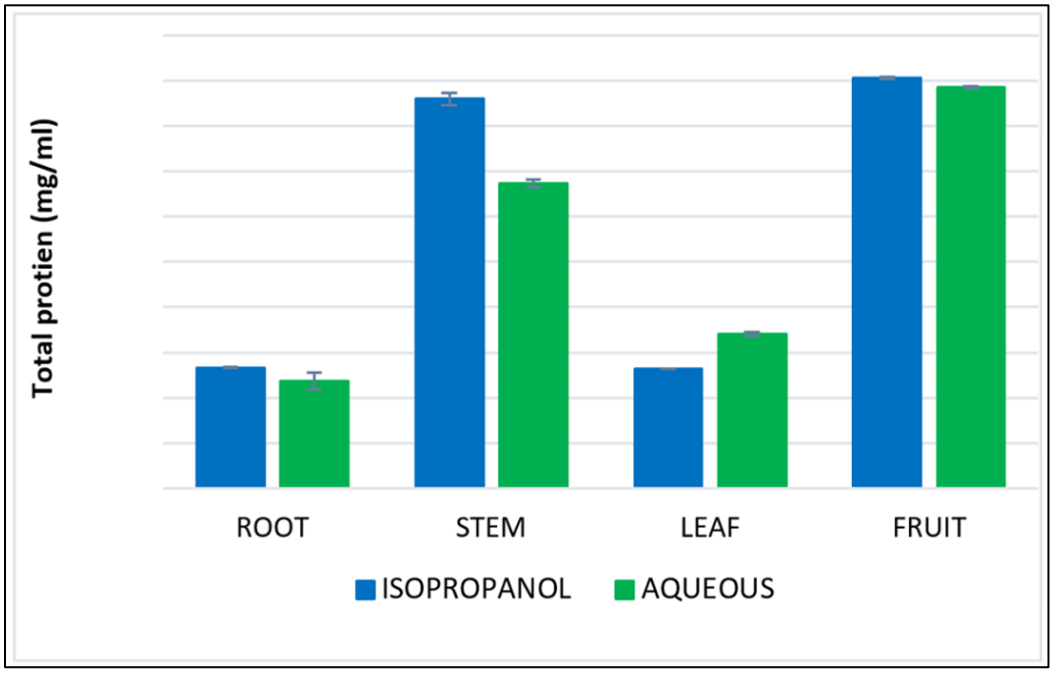

Figure.6 Quantification of total protein from extracts of *E. hirta*

DISCUSSION

This study declares that *E. hirta* possess the novel phytochemicals and many other antioxidant compounds which help to prevent the oxidative stresses and various diseases. These phytochemicals have potential to resist against the microbes. The phenolic and flavonoids compound are very effective to resist microbes (Al-Snafi, 2017). Due to the presence of these compounds in *E. hirta*, it has high medicinal value.

Antibacterial activity of extracts of *E. hirta* were revealed by determining the diameters of zones of growth inhibition against two bacterial species of Gram negative (*E. coli* and *K. pneumonia*) and one Gram positive (*S. aureus*) as presented in Table-1. All tested bacterial species were susceptible to *E. hirta* extracts in variable degree. It was described that plant extracts are effective to bacteria on the basis of different degree of zone of inhibition for types and species. (Davis & Stout, 1971) have described the criteria for antimicrobial power based on the diameter of the inhibitory zone. According to them if the inhibitory zone < 5mm considered in the weak category, between 5-10 mm in the medium category and > 20 mm categorized as very Strong. It was reported that the extracts of the *E. hirta* herb contains phytochemicals that act as antimicrobials (Simanjuntak & Rahmiati, 2021).

The results presented in this study revealed that most of the isopropanol and aqueous extracts of *E. hirta* root, stem and fruit against *S. aureus* displayed negative results that was proved by previous work (Alisi & Abanobi, 2012) in which it has been described, ethanol extract of *E. hirta* was ineffective towards *S. aureus.* However aqueous leaf extract produced large inhibition zone (18 mm) whereas small inhibition zone (10 mm) by isopropanol fruit extract as compared to standard Avelox (12 mm), Erythromycin (15 mm) and Ofloxacin (14.6 mm). These results are lower than those of (Rajeh, Zuraini, Sasidharan, Latha, & Amutha, 2010) who have calculated positive results in term of zone of inhibition from the range of (16 to 28 mm) in all methanol extracts of root, stem, leaf and fruit of *E. hirta* against *S. aureus.* On the other hand the reported result is higher in present study than those of (Anarado, Anarado, Umedum, Chukwubueze, & Anarado, 2020) who have reported that methanol extract of *E. hirta* leaf showed highest inhibition zone towards *S. aureus* was 9.93 mm while resistant towards n-hexane and ethyl acetate. (K. R. SHARMA, 2020) has also reported that methanol extract of *E. hirta* plant sensitive towards *S. aureus* with zone of inhibition 10 mm. Surprisingly, (Tran, Nguyen, *et al.*, 2020) have revealed that all extracts of *E. hirta* i-e methanol, butanol, ether, chloroform and ethyl acetate showed resistant towards *S. aureus and E.coli.*

On the other hand, regarding the present report, the highest inhibition zone was shown by isopropanol stem extract (25 mm) against *E. coli.* This result is in good agreement with the earlier study (Pandey & Verma, 2013), but the solvent was different the methanolic leaf extract of *E. hirta* revealing the maximum zone of inhibition (25 mm) against *E. coli*. However, the lowest zone of inhibition was shown by aqueous root and fruit (11.3 mm) in *E. hirta* against *E. coli*. Generally, our results are higher than previously reported work of (Rajeh *et al.*, 2010), who have reported that highest zone of inhibition (18 mm) from methanol leaf extracts of *E. hirta,* it was reported (Anarado *et al.*, 2020) that ethyl acetate extract of *E. hirta* leaf showed highest inhibition zone towards *E. coli* was 0.624 mm while N-hexane extract showed minimum zone of inhibition 0.173 mm and resistant towards methanol extract, (K. R. SHARMA, 2020), has reported that methanol extract of *E. hirta* plant sensitive towards *E. coli* with zone of inhibition 10 mm and (Simanjuntak & Rahmiati, 2021) have reported that various concentrations of ethanol extract of *E. hirta* plant showed variable zone of inhibition towards *E .coli* from 10 to 31 mm. Overall it was concluded from current report that all extracts of *E. hirta* produced effective zone of inhibition but maximum zone of inhibition was showed in isopropanol stem extract of *E. hirta* as compare to standard (antibiotics Erythromycin :18 mm) against *E. coli*.
Furthermore, *K. pneumonia* was used to check the antibacterial activity of *E. hirta* in present study. The large size of zone of inhibition was produced in isopropanol root extract (10.75 mm) but low size of zone of inhibition was produced in aqueous fruit (6 mm). Overall it was observed that all extracts showed decreased inhibition zone than standard Avelox and Erythromycin (20 mm) towards *K. pneumonia.*

In present work quantity of phytochemicals were examined and results were obtained that the highest antioxidant activity was obtained 0.418 mg/ml in aqueous fruit extract and lowest antioxidant activity was analyzed 0.222 mg/ml in isopropanol root extract of *E. hirta*. The presented results are quite higher than previously reported work of (Basyal, Neupane, Pandey, & Pandeya, 2021) who have investigated the antioxidant activity of ethyl acetate extract of *E. hirta plant* which was found to be 0.032 mg/mL, (Tran, Nguyen, *et al.*, 2020) have reported that all extracts of *E. hirta* i-e methanol (0.017), butanol (0.056), ether (0.123), chloroform(0.092) and ethyl acetate(0.01 mg/ml). (K. R. Sharma, 2020) has reported that methanol extract of *E. hirta* plant have antioxidant activity 0.029 mg/ml. Overall it was observed that very high antioxidants contents were present in all parts of *E. hirta* plant extracts that was proved by earlier work of (Rajeh *et al.*, 2010) who have calculated antioxidant activity from methanol extracts of root, bark, leaf and flower of *E. hirta* plant.

However highest and lowest phenolic contents in the *E. hirta* plant were investigated as 0.176 mg/ml in aqueous leaf and 0.086 mg/ml in aqueous root respectively. Previous work also has been reported that leaf extracts had the highest total phenolic (Huang, Chen, & Yang, 2012). Our results are quite lower than those of (Basyal *et al.*, 2021) who have prepared ethyl acetate extract of *E. hirta* plant and the phenolic contents was found to be 288.10 mg GAE/g. (Aqib, 2021) has estimated methanol extract of leaf of *E. hirta* 0.0023 mg/100g, (Anarado *et al.*, 2020) have reported that ethyl acetate extract of *E. hirta* leaf contained total phenolic 8.59 mg/l, (Tran, Nguyen, *et al.*, 2020) have estimated that all extracts of *E. hirta* i-e methanol 110, ether 91, chloroform 56, ethyl acetate 255 and butanol 71 mg GAE/g phenolic and (K. R. SHARMA, 2020) has analyzed that methanol extract of *E. hirta* plant contained phenolic 138 mg GAE/g.

In flavonoids determination the maximum value was obtained 0.48 mg/ml in aqueous leaf extract and minimum value was investigated 0.17 mg/ml in isopropanol root extract from *E. hirta* plant. The presence of these bioactive compounds recovered in present study from the traditional medicinal plant (*E. hirta*) was found to cease the growth of some Gram positive and Gram negative bacteria. Other reports had also revealed that medicinal plants contain phenolics, flavonoids, coumarins, alkaloids, tennins, terpenoids and polyacetylenes which have the potential as a bacteriostatic, bactericidal or fungicidal efficacy versus certain human pathogens (Dholaria & Desai, 2018; Motaleb, 2011).

The total sugar, reducing sugar and total protein were recorded in significant amount in all parts of *E. hirta* extracts. It was noted that there are very few reports are available in literature regarding quantification of biomolecules from *E. hirta* plant.

According to present study, the presence of phytochemicals was analyzed qualitatively and quantitatively that were found in aqueous and isopropanol root, stem, leaf and fruit extracts of *E. hirta* in significant amount. Many researchers have also been evaluated these different phytochemicals in variable amounts (Islam *et al.*, 2013) (Abubakar, 2009; Alisi & Abanobi, 2012; Roy, Rao, Bhuvaneswari, Giri, & Mangamoori, 2010).

CONCLUSION

It is concluded that *E. hirta* contains phytochemicals and biochemicals such as alkaloids, steroids, glycosides, flavonoids, tannins, coumarin, phenolics, carbohydrates and proteins in varying concentrations. These constituents were isolated from aqueous and isopropanol extracts of different parts of *E.*

hirta. It is also concluded that the *E. hirta* were highly effective to growth inhibition against *E. coli, S. aureus and K. pneumonia*. It is suggested that different parts of this plant has great potential efficacy against different human pathogenic diseases.

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