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Evaluation of *in vitro* antioxidant activity of different solvent extracts from *Grewia hirsuta* (Vahl)

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Abstract

Antioxidant provides protection to human against various degenerative diseases as it plays an important role in scavenging free radicals. Due to the safe therapeutic values of various medicinal plants now the modern research is more directed towards "Natural antioxidants". *G. hirsuta* belonging to the family Tiliaceae has high medicinal values in traditional medicine. In the present study antioxidant activity of leaf and fruit extracts of *Grewia hirsuta* were carried out. Results revealed that both the extracts have sound scavenging activity.

Keywords: Free radicals, DPPH, Antioxidant activity, *Grewia hirsute*

Introduction

Oxidative Stress (OS) is a general term used to describe the steady state level of oxidative damage in a cell, tissue or organ, caused by the reactive oxygen species (ROS). Reactive oxygen species (ROS), such as superoxide anions, hydrogen peroxide, hydroxyl, nitric oxide and peroxy nitrite radicals play an important role related to the pathogenesis of various important diseases (Bharti *et al*., 2012 and Ansari *et al*., 2013).Antioxidants refer as a broad range of compounds which have ability to neutralize free radicals by donating one of their electrons. It acts in different ways by preventing the propagation of the oxidative chain reaction by scavenging free radicals by being part of redox antioxidant network, or by regulating gene expression (Ahmad *et al*., 2013). It is believed that natural antioxidants rich food can actually lower risks of degenerative diseases and prevent oxidative stress therefore recently many medicinal plants have been studied for their antioxidant properties (Elkhamlichi *et al*., 2017 and Chen *et al*., 2013). Keeping this in view, antioxidant activity of leaf and fruit extracts of *Grewia hirsute* by DPPH assay were carried out. *G. hirsuta* belonging to the family Tiliaceae has high medicinal values in traditional medicine such as diarrhea and dysentery, heart diseases, wounds, cough, swellings, dyspnea (Ema *et al*., 2013, Jeyaprakash *et al*., 2011).

Materials and Methods

Collection of plant material

The fresh sample of leaf and fruit of *G.hirsuta* (Fig.-1) were collected from various parts of Buldhana district, Maharashtra and identified with the help of floras (Cooke, 1903; Dhore, 1986 and Kirtikar and Basu, 1995). Collected material was air dried under shade. After drying, the plant material was ground well using mechanical blender into fine powder and transferred into airtight containers with proper labeling.

Fig.-1 ***Grewia hirsuta*** **(Vahl)**

Preparation of the plant extracts

Crude plant extracts were prepared by Soxhlet extraction method. About 50 gm of dried powdered material of leaf and fruit was uniformly packed into a thimble and extracted with 500 ml of different solvents separately. Solvents used were ethanol, ethyl acetate, acetone, chloroform and distilled water. The process of extraction continues for 24 hours or till the solvent in siphon tube of an extractor become colorless. All the extracts were evaporated to dry and dried extracts were kept in refrigerator at 4ºC for their future uses.

Antioxidant activity with DPPH assay

The ability of the plant extracts to scavenge DPPH free radical was assessed by standard method (Blois, 1958). Five concentrations (25, 50, 75, 100ug/ml) of each sample were prepared. 0.1 mM solution of DPPH in methanol was prepared and 180 µl of this solution was added to 20µl of different plant extracts in 96 well plates and incubated. After 30 min incubation in dark at room temperature the absorbance was recorded at 490 nm. Ascorbic acid was used as a positive control. Percentage inhibition was calculated using following equation, while IC50 values were estimated from the % inhibition versus concentration plot. The effective concentration of sample required to scavenge DPPH radical by 50% (IC50 value) was obtained by linear regression analysis of dose response curve plotting between % inhibition and concentrations. The absorbance was recorded and % inhibition was calculated using formula given below, (Badami and Gupta, 2005).

$$I\ \% = (Ac - As) / Ac \times 100 \ldots\ldots (1)$$

Where, Ac – absorbance of the control
As – absorbance of the sample

Results and Discussion

Table- 1: Evaluation of DPPH radical scavenging activity of *Grewia hirsuta* (Leaf and Fruit)

Extracts	Concentration (µg/ml)	Leaf		Fruit	
		%Inhibition (Mean± SD) n=2	IC50 (µg/ml)	%Inhibition (Mean± SD) n=2	IC50 (µg/ml)
Ethanol	25	51.74 ±1.58	28.17	42.64±1.76	49.96
	50	52.61±4.23		52.49±2.64	
	75	54.61±2.11		53.49±1.58	
	100	66.58±3.87		65.46±2.64	
Ethyl acetate	25	49.25±2.64	34.72	46.00±1.93	46.55
	50	51.37±1.76		49.87±4.23	
	75	52.61±1.05		54.73±2.29	
	100	54.86±1.05		66.45±6.17	
Acetone	25	27.55±0.52	100.31	51.37± 2.11	13.72
	50	35.53±3.70		61.47±2.29	
	75	45.88±2.11		63.96±2.64	
	100	48.00±1.23		70.69±1.93	
Chloroform	25	35.53±1.23	85.93	45.26±5.46	47.52
	50	40.77± 2.29		51.99±3.35	
	75	46.75±2.29		53.11±3.52	
	100	54.11±2.11		62.09±2.46	
Aqueous	25	44.26±0.17	33.94	41.02±5.11	58.74
	50	58.47±0.88		50.62±2.11	
	75	63.21±0.52		54.48±2.99	
	100	65.58±0.46		56.96±0.88	
Ascorbic acid	25	52.86±3.87	9.39	52.86±3.87	9.39
	50	63.96±2.64		63.96±2.64	
	75	74.31±1.76		74.31±1.76	
	100	75.56±2.82		75.56±2.82	

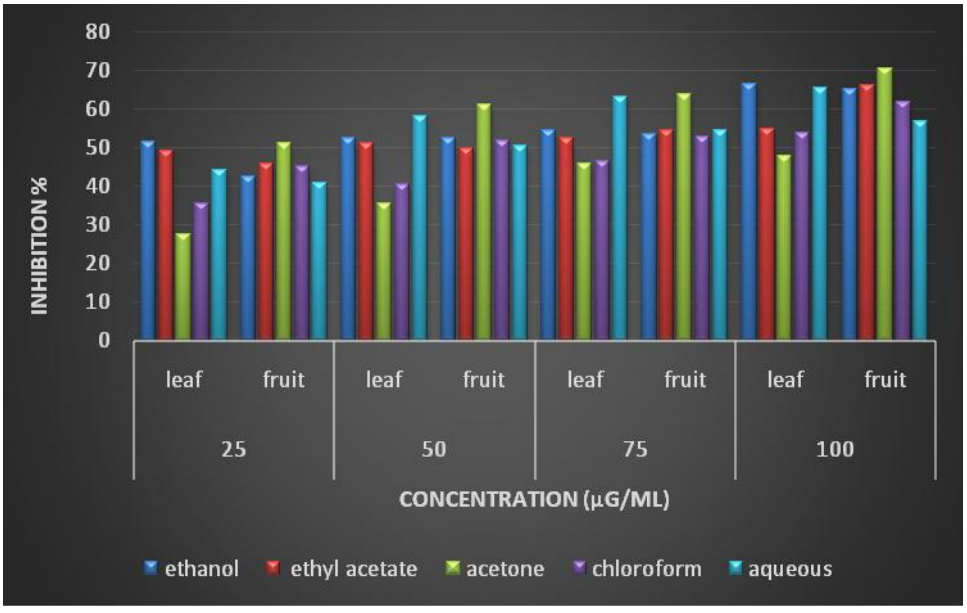

Figure-2. Antioxidant activity of *Grewia hirsuta* (Leaf and fruit) extracts

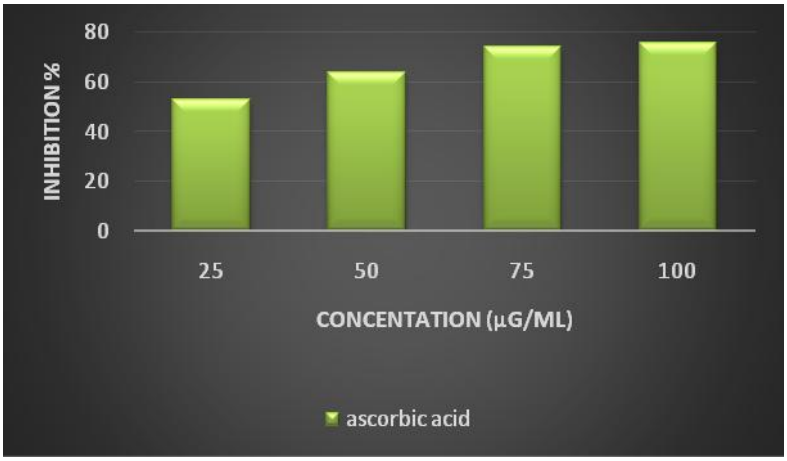

Figure-3. Antioxidant activity of Ascorbic acid

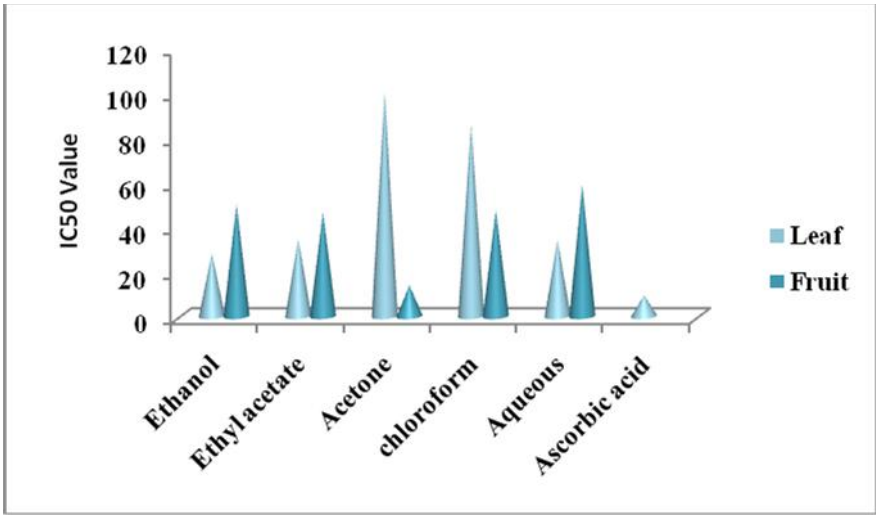

Figure-4: IC50 values of *Grewia hirsuta* (Leaf and Fruit) extracts

DPPH model is a simple, precise, relatively quick and acceptable method which is widely used to measure free radical scavenging activity. It has been employed in the determination of antioxidant ability of numerous natural products (Ogunva *et al*., 2016). In the present investigation antioxidant activity *G. hirsuta* (Leaf and fruit) was studied with DPPH assay. Ethyl acetate, acetone, ethanol, chloroform and water extracts were used to analyze antioxidant property and ascorbic acid was used as standard (Fig. 3). Results are incorporated in Table-1.The study revealed that *G. hirsute* (Leaf and fruit) possesses sound antioxidant activity. The acetone extract of the fruit showed highest antioxidant activities (70.69±1.93) followed by ethyl acetate, ethanol, chloroform and aqueous extracts (Fig 2). It also showed minimum IC50 value i.e. 13.72µg/ml which was nearer to IC50 of ascorbic acid 9.39µg/ml (Fig. 4). The ethanol fraction of the leaf sample showed 66.58±3.87 % scavenging activity with IC50 value i.e. 28.17µg/ml followed by aqueous 65.58±0.46% scavenging and 33.94 µg/ml IC50. In both the samples free radical scavenging activity was found to increase with the increase in the concentration of the fractions. There is less reports are available on the antioxidant activities of *G. hirsuta*. However, Anwar *et al*., (2015) had reported the antioxidant activity of *Grewia optiva* and Hamid *et al*., (2016) in *G. pubescens*. Thirugnanasambandan and Kannayiram (2016) reported that acetone extract of fruit of *Grewia umbelliferes* exhibited significant DPPH free radical scavenging activity with significant IC50 value. Meena *et al*., (2014) reported aqueous extract of *G. nervosa* was less effective in antioxidant property. Kshirsagar and Upadhyay (2009) reported the antioxidant activity in *G. nervosa* and *G. sapida* and Arora (2011) in *G. optiva*. Hutke and Naswale (2019) reported that ethyl acetate extract showed maximum antioxidant activity *G. tiliaefolia.*

Conclusion

In the present study antioxidant activity *G. hirsuta* leaf and fruit extracts were evaluated. The acetone extract of fruit showed maximum antioxidant activity.

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