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Comparative Evaluation of *in vitro* Free Radical Scavenging Activity of Different Extract of *Salvadora persica L.*

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Abstract:

Salvadora persica L. is a plant with opposite branches widely cultivated in farms and along street as wind barriers. Evaluation of in vitro free radical scavenging activity of different extracts of Salvadora persica L. was performed. Petroleum ether, ethanolic and aqueous extracts of Salvadora persica L were prepared in a soxhlet apparatus. Each extract was selected to study the free radical scavenging activity by superoxide scavenging assay method (DPPH assay). It was found that the aqueous extract contained carbohydrates, glycosides, flavonoids, alkaloids and steroids. The ethanolic extract contained glycosides, flavonoids, alkaloids and steroids. The ethanolic extract of Salvadora persica L. showed 65.99 ± 0.45% inhibition in the superoxide scavenging model. The aqueous extract also showed comparable activity (55.22 ± 0.61%), while the petroleum ether extract showed poor superoxide scavenging activity. Both extracts showed the dose and time-dependent inhibition in the DPPH scavenging model.

Keywords: *DPPH scavenging, antioxidant activity, Salvadora persica L., medicinal plants.*

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INTRODUCTION

Salvadora persica L. is a large shrub with opposite branches, sometimes growing as dense thickets on sand hummock, belonging to family Salvadoraceae, commonly known as 'Pilu', 'Jal' and 'Tooth brush tree' and is widely distributed in India, Africa, Saudi Arabia, Iran, Israel and Pakistan. It has been claimed in traditional literature to be valuable against a wide variety of diseases [1-3]. The plant is mentioned in holy Quran and Sonnah, the roots and shoot sticks have been used for centuries as oral hygiene tools in many parts of the world. It was reported that fresh and dried leaves, dried fruits and stems are used to treat swellings, ulcers and blisters, scorpion stings, regulating menstruation, gases and worms. In many parts of the world roots used as toothbrushes and the crushed leaves used with oil to treat joint and knee pains. The fruits are edible and used as a carminative, anthelmintic, vulnerary, stomachic, antiseptic and anti inflammatory. Leaves and flowers also used for toothache, gumproblems, skin diseases, kidney stones, constipation and anthelmentic. Also the plant has been incorporated into commercially available toothpaste.

Salvadora persica L. extract showed presence of carbohydrates, alkaloid (salvadorine), steroids, terpenoids, saponins, flavonoids (quercetin & kaempferol) and glycosides.[4-7].

Free radicals are atoms or groups of atoms with an odd number of electrons and can damage important cellular components such as DNA or cell membranes. To prevent free radical damage, the body has a defense system of antioxidants [8]. Scientific literature reveals that the carbonyl groups present in the flavonoids and phenolic compounds are responsible for the free radical scavenging activity [9]. Literature survey reveals that flavonoids and other phenolic compounds are present in plant *Salvadora persica* L. which are known to be responsible for the antioxidant activity; since it has phytoconstituents of antioxidant interest, thus present research concluded comparative evaluation of antioxidant activity of different extracts of *Salvadora persica* L..

MATERIALS AND METHODS

Collection of leaves of the *Salvadora persica* L. was done from Jodhpur, India. 1,1-diphenyl 2-picryl hydrazyl (DPPH) was purchased from Sigma Aldrich. All other chemicals and reagents were of analytical grade.

Preparation of the extracts:

The leaves of *Salvadora persica* L. were collected and shade dried. The dried leaves were coarse powdered and the powder was packed into a soxhlet column and extracted successively with petroleum ether (60–80^{0}C), ethanol and with distilled water. The extracts were concentrated under reduced pressure. The dried extracts were stored in an airtight container in a refrigerator below 10–20^{0}C.

Preliminary phytochemical screening:

The preliminary phytochemical screening was carried out on petroleum ether, ethanolic and aqueous extracts of *Salvadora persica* L. Tests for common phytochemicals were carried out by standard methods [10, 11].

Superoxide scavenging activity:

Petroleum ether, aqueous and ethanolic extracts of leaves of *Salvadora persica* L. were screened for anti-oxidant activity using superoxide free radical scavenging model in a dose and time-dependent manner [12]. The assay was based on the capacity of the samples to inhibit blue formazan formation by scavenging the superoxide radicals. The DPPH radicals are reduce to corresponding hydrazine when

it react with hydrogen donors, the DPPH radical is purple coloured compound & upon reaction with hydrogen donors it becomes colourless. It is discolouration assay which is evaluated by the addition of the sample drug (antioxidant) to DPPH solution in methanol & the reduction in absorbance was measured on 517 nm. The reaction mixture was contained 0.1 mM DPPH with methanol & different concentrations of (10–100 µg/ml) of samples for the incubation period of 30 min. Mixture of equal volume of methanol & DPPH was used as control. Ascorbic acid was used as a standard drug. Percentage inhibition and IC_{50} values were calculated (Results are shown in Table 1, Fig. 1).

RESULTS & DISCUSSION

It was found that the petroleum ether extract contained steroids, fat and fixed oils. Aqueous and ethanolic extracts contained carbohydrates, alkaloid (salvadorine), steroids, terpenoids, saponins, flavonoids and glycosides. The ethanolic extract of *Salvadora persica* L. showed 65.99 % inhibition in the superoxide scavenging model at the concentration of 50 µg mL $^{-1}$. Activity decreased for aqueous extract; 55.22 % compared to the ethanolic extract at the same concentration 50 µg mL $^{-1}$ (Fig. 1.), while the petroleum ether extract showed poor superoxide scavenging activity.

All the extracts showed dose and time-dependent inhibition of the superoxide scavenging activity. The results are reported in Table 1.

Statistical analyses:

Data are the mean ± SD of three measurements. Statistical analysis was performed by the Student's t-test and by ANOVA.

CONCLUSION

In this investigation *in vitro* free radical scavenging activity of different extracts of *Salvadora persica* L. were performed comparatively, it was found that ethanolic and aqueous extracts of leaves of the *Salvadora persic*a L. showed significant free radical scavenging activity; it was suggested that presence of flavonoids and other phenolic compounds may be responsible for the free radical scavenging activity of leaves extract of plant. Ethanolic extract showed most potent antioxidant activity as compared to other extracts. Thus research concluded that ethanolic extract of *Salvadora persica* L. can be used as potent antioxidant which can play vital role against the disease i.e; cancer. Potent antioxidant activity of *Salvadora persica* L. increased its width as useful traditional medicine.

Table 1. % Inhibition of different extracts of *Salvadora persica* L.

S.No.	Concentrations (µg mL $^{-1}$)	% Inhibition		
		Petroleum ether Extract	Ethanolic Extract	AqueousExtract
1.	10	22.9±0.12	39 ± 0.36	33.8 ± 0.29
2.	20	27.0±0.17	44.45 ± 0.54	38.94 ± 0.65
3.	30	31.8±0.29	49.29 ± 0.61	44.37 ± 0.44
4.	40	37.0±0.31	55.19 ± 0.67	49.23 ± 0.67
5.	50	41.4±0.38	65.99 ± 0.45	55.22 ± 0.61
6.	100	47.6±0.41	72.45 ± 0.39	63.12± 0.65

Note: Data are the mean ± SD of three measurements.

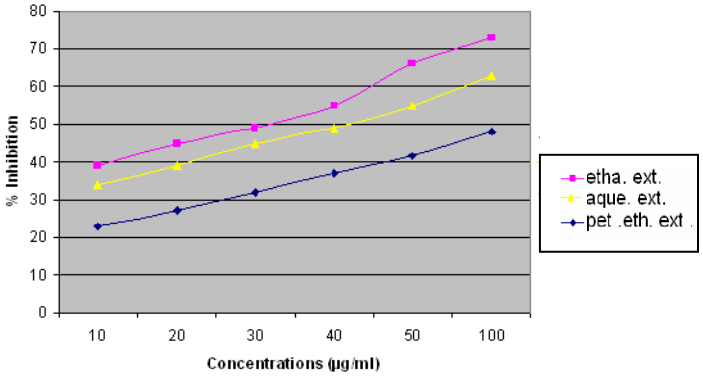

Fig. 1. % Inhibition of different extracts of *Salvadora persica* L. (DPPH Assay)

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