

## COMPARATIVE EVALUATION OF FREE RADICAL SCAVENGING ACTIVITY OF *Cleome viscosa* AND *Trichodesma indicum*

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### ABSTRACT

**Objective:** The present study was attempted to compare the *in vitro* free radical scavenging activity in the leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum*.

**Methods:** The leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum* were extracted with solvents namely petroleum ether, benzene, chloroform, ethyl acetate and ethanol for the determination of free radical scavenging activity. The antioxidant activity was evaluated by determining ABTS, DPPH, superoxide, nitric oxide, hydroxyl radicals, hydrogen peroxide scavenging and inhibition of *in vitro* lipid peroxidation.

**Results:** The chloroform and ethyl acetate extract of the roots of *Trichodesma indicum* and the ethyl acetate extract of the fruit of *Cleome viscosa* exhibits strong inhibition against DPPH radicals. The ethyl acetate and ethanol extract of the leaves, fruits and roots of *Cleome viscosa* and ethanol extract of the leaves, fruits and roots of *Trichodesma indicum* showed significant ABTS radical scavenging activity. Petroleum ether extract of the leaves of *Cleome viscosa* and *Trichodesma indicum* was found to possess potent superoxide scavenging activity. The maximum nitric oxide scavenging activity was found to be exhibited by the benzene extract of the fruit of *Trichodesma indicum*. Ethyl acetate and benzene extract of the leaf of *Trichodesma indicum* reveals the significant activity of hydroxyl radical scavenging activity. **Conclusion:** Based on the results, it can be concluded that the extracts of both the medicinal plants *Cleome viscosa* and *Trichodesma indicum* have great potential of free radical scavenging activity.

**Keywords:** *Cleome viscosa*, *Trichodesma indicum*, DPPH, ABTS.

### INTRODUCTION

Free radicals are chemical species, which contains one or more unpaired electrons due to which they are highly unstable and cause damage to other molecules by extracting electrons from them in order to attain stability [1]. The biological reactivity of free radicals and their roles in oxidative stress are subjects of considerable attention and controversy. Oxygen consumption, necessary for cell growth, generates reactive oxygen species (ROS). The body's normal metabolism, normal use of oxygen, such as respiration and some cell-mediated immune functions continuously produce ROS. Being active in biological systems, superoxide anion radical ( $O_2^{\bullet-}$ ), hydrogen peroxide ( $H_2O_2$ ), hydroxyl radical ( $\bullet OH$ ), and other reactive oxygen species are collectively referred to as ROS. They are capable of attacking the healthy cells of the body, causing them to lose their structure and function, if cellular agents do not effectively scavenge ROS, then disease will ensue and they are implicated in more than 100 diseases. Cell damage caused by free radicals appears to be a major contribution to aging and degenerative diseases of aging such as cancer, cardiovascular disease, cataracts, immune system decline, liver diseases, diabetes mellitus, inflammation, renal failure, brain dysfunction and stress among others. To protect the cells and organ system of the body against reactive oxygen species, human have evolved a highly sophisticated and complex antioxidant protection system that functions interactively to neutralize free radicals. Thus, antioxidants are capable stabilizing or deactivating, free radicals before they attack cells [2]. Many natural products are reported to contain large amounts of antioxidants [3].

Cellular defenses against ROS include enzymes such as catalase which degrades hydrogen peroxide into water and oxygen and antioxidants such as tocopherol and ascorbic acid neutralize them. Most of these antioxidants in cells and tissues are supplied through the diet [4].

Plants constitute an important source of active natural products which differ widely in terms of structures, biological properties and mechanisms of actions. Various phytochemical components are known to be responsible for antioxidant, antimicrobial, antilarvicidal and anti-inflammatory activities of plants.

*Cleome viscosa* L., (Cleomaceae) is a widely distributed sticky herb with yellow flowers and long slender pods containing seeds, which

similar to those of mustard (Hindi), Hurhuria (Bengali), Nayikkadugu (Tamil) in South Asian folk medicine, found throughout the larger part of Indian Subcontinent, often in waste places [5].

*Trichodesma indicum* (Adhaphushpi) is a perennial medicinal herb distributed in tropical and subtropical Asia, Africa and Australia. It is a multidrug plant used to reduce or cure inflammation, pain, osteoarthritis and conjunctivitis. This plant is used for expulsion of dead fetus abortion, inhibition of diarrhoea, reduction of sulfur dioxide-induced cough reflex in mice, as brain tonic and to treat breast cancer [6].

With this background, the present study was formulated to assess and compare the free radical scavenging activity of *Cleome viscosa* and *Trichodesma indicum*.

### MATERIALS AND METHODS

#### Collection of plant material

The plant samples were collected from Erode district and were identified by the Botanical Survey of India, Tamil Nadu Agricultural University Coimbatore. Fresh leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum* (Plate I and II) were collected and cleaned to remove adhering dust particles, washed under running tap water and gently blotted dry between folds of tissue paper. They were then dried at room temperature in shade away from direct sunlight, powdered and stored at room temperature until use.

#### Chemicals

ABTS (2,2'-azino-bis-3-ethyl benzothiazoline-6-sulphonate), hydrogen peroxide, glacial acetic acid, n-propanol, pyridine, thiobarbituric acid, deoxyribose, sodium nitroprusside, riboflavin, ethylene diamine tetra-acetic acid (EDTA), 1,1-diphenyl-2-picrylhydrazyl (DPPH) were obtained from Himedia and Merck. All the other chemicals and solvents used were of analytical grade.

#### Quantification Assays

##### Preparation of the plant extracts

The leaf, fruit and root powder of *Cleome viscosa* and *Trichodesma indicum* (10g) was extracted with 100ml of various

solvents namely petroleum ether, benzene, chloroform, ethyl acetate and ethanol. The organic extracts were obtained by using Soxhlet apparatus. Period of extraction was 24 hours. Then each extract was concentrated to dryness under pressure to obtain the dry extracts. They were then dissolved in dimethyl sulfoxide (DMSO) and stored in the refrigerator until use. 10mg of extracts per 20 $\mu$ l was taken for the assay.

Plate I: *Cleome viscosa*

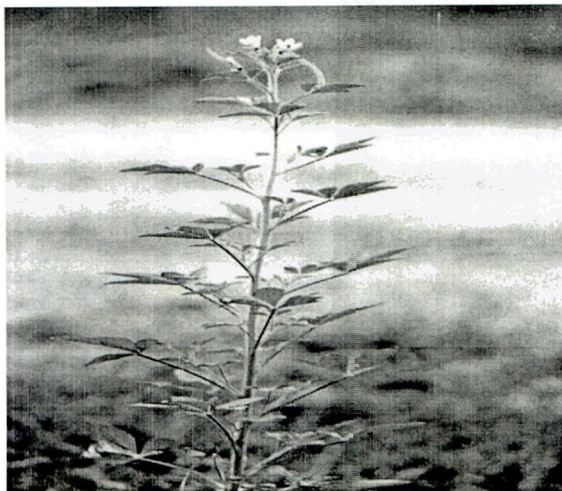


Plate II: *Trichodesma indicum*



#### ABTS radical scavenging effects

The ABTS radical scavenging activity was measured by the method of Shirwaikar *et al.* [7].

ABTS (2,2'-azino-bis-3-ethyl benzthiazoline-6-sulphonic acid) radical cation de-colourisation assay was employed to assess the radical-scavenging effect of the leaf extracts of the candidate plant. ABTS is a chromogen, which changes into a coloured mono-cation radical form (ABTS<sup>•+</sup>) in the presence of oxidative agent and the ABTS<sup>•+</sup> has an absorption peak at 750nm. Antioxidants will reduce ABTS<sup>•+</sup> into its colourless form and the extent of decolourisation corresponds to the per cent reduction of ABTS. The six different extracts (100 $\mu$ l each) were added to ABTS solution (300 $\mu$ l) and the final volume of each was made up to 1ml with ethanol. The absorbance was read at 745nm and the percentage inhibition was calculated using the formula,

$$\text{Inhibition (\%)} = \frac{A(\text{Control}) - A(\text{Sample})}{A(\text{Control})} \times 100$$

#### DPPH radical scavenging activity

The extracts of leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum* were checked for their antioxidant property using the biological end point of *in vitro* DPPH scavenging activity by the method of Mensore *et al.* [8]. Antioxidants react with DPPH and convert it to diphenyl-picryl hydrazine by donating its OH group. The degree of discoloration from purple to yellow colour can be measured at 519nm, which is a measure of the radical scavenging potential of the extracts. The different solvent extracts and crude aqueous extract (5 $\mu$ l) was added with 0.5ml of methanolic solution of DPPH and 0.495ml of methanol. The mixture was then allowed to stand at room temperature for 30 minutes. DPPH methanol solution was used as positive control and methanol alone acted as blank. After incubation, the conversion of purple colour to yellow colour was read at 518nm in a spectrophotometer. The per cent inhibition was calculated using the following formula;

$$\text{Scavenging activity (\%)} = \frac{100 - A(\text{Control})}{A(\text{Control}) - A(\text{Sample})} \times 100$$

#### Superoxide scavenging activity

Inhibition of superoxide generation was estimated by the method of Winterbourn *et al.* [9]. The extent of superoxide generation was studied on the basis of inhibition of the production of nitrobluetetrazoliumformazon of the superoxide ion by the plant extracts and is measured spectrophotometrically at 560nm. The assay mixture contained 0.02ml of plant (solvent and crude aqueous) extracts with 0.2ml of EDTA, 0.1ml of NBT, 0.05ml of riboflavin and 2.63ml of phosphate buffer. DMSO, instead of plant extract, was considered as control. All tubes were vortexed and the initial absorbance was read at 560nm. The tubes were illuminated uniformly using a fluorescent lamp for 30 minutes. The absorbance was read again at 560nm. The difference in optical density before and after illumination is the measure of superoxide generation and the percentage inhibition was calculated using the formula,

$$\% \text{ Superoxide Scavenging} = \frac{A(\text{After illumination}) - A(\text{Reference})}{A(\text{Control})} \times 100$$

#### Nitric oxide scavenging activity

Inhibition of nitric oxide generation was determined by the method of Green and Hill [10]. An aqueous solution of sodium nitroprusside spontaneously generates nitric oxide at physiological pH, which interacts with oxygen to produce nitrite ions, which is measured at 546nm. Sodium nitroprusside (2.0ml), phosphate buffered saline (0.5ml) and each of the six different plant extracts (0.5ml) were mixed and incubated at 25°C for 30 minutes. Griess reagent (0.5ml) was added and allowed to stand for 30 minutes. The control tube was prepared without leaf extracts. The absorbance of the pink coloured chromogen was read at 546nm against a reagent blank.

$$\text{Nitric oxide scavenging activity (\%)} = \frac{A(\text{Sample})}{A(\text{Control})} \times 100$$

#### Hydroxyl radical scavenging effects

The hydroxyl radical scavenging activity was measured by the method of Elizabeth and Rao [10]. The reaction mixture contained deoxyribose (2.8Mm), EDTA (0.1Mm) and potassium dihydrogen phosphate - potassium hydroxide buffer (20Mm, pH<sup>7.4</sup>). 20 $\mu$ l of sample was added such that the final volume was 1.0 ml. The reaction mixture was incubated for 1 hour at 37°C. Deoxyribose degradation was measured as TBARS by adding 0.5ml of TBA and 0.5 ml of HCl, boiled in a water bath for 20 min, cooled and measured the absorbance at 532nm.

#### Hydrogen peroxide scavenging activity

The ability of the plant extract to scavenge H<sub>2</sub>O<sub>2</sub> was determined according to the method of Ruchet *et al.* [11]. A solution of H<sub>2</sub>O<sub>2</sub> (4mM) was prepared in phosphate buffer (pH 7.4) concentration was determined spectrophotometrically from its absorption at 230 nm was determined after 10 minutes against a blank solution containing phosphate buffer without H<sub>2</sub>O<sub>2</sub>.

The scavenging activity of  $H_2O_2$  by plant extract and the standard compounds was calculated using the formula.

$$\% \text{ Scavenging activity} = \frac{A_0 - A_1}{A_0} \times 100$$

$A_0$  - Absorbance of control

$A_1$  - Absorbance of sample at 230 nm

#### Inhibition of *in vitro* lipid peroxidation

Quantitative measurement of lipid peroxidation in goat liver homogenate was performed by the method of Okhawa *et al.* [13]. The reaction mixture containing 0.1ml each liver homogenate, plant extract (50 $\mu$ l), 50 $\mu$ l ferrous sulphate and add TBS to make a final volume to 500 $\mu$ l. A blank containing no lipid source but only ferrous sulphate, no plant extract and TBS to final volume of 0.5ml was prepared.

As assay medium corresponding to 100% oxidation was prepared by adding all the other constituents except plant extract. The experimental medium corresponds to autooxidation contained only of liver homogenate. Tubes are incubated at 37°C for 1 hr. After incubation, 0.5ml of 70% ethanol was added to all tubes to arrest the reaction. 1ml of 1% TBA was added to all the tubes. The tubes were then incubated in a boiling water bath for 20 minutes. After cooling room temperature, added with 0.5ml of acetone. The intensity of pink colour produced is measured at 535nm in a spectrophotometer.

#### Statistical analysis

All experimental data were expressed as mean  $\pm$  SD. The results were analyzed by two way analysis of variance (ANOVA). Significant difference was accepted at the  $p < 0.05$  level.

#### RESULTS AND DISCUSSION

Medicinal plants constitute one of the main sources of new pharmaceuticals and health care products. A whole range of plant

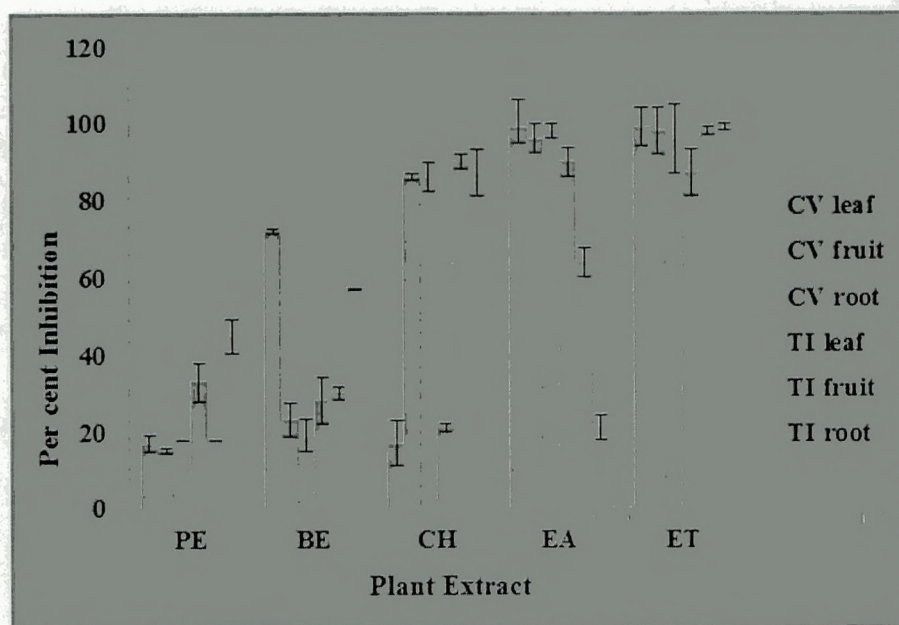
derived dietary supplements, phytochemicals and pro-vitamins that assist in maintaining good health and combating disease are now being described as functional ingredients and nutraceuticals. The role of medicinal plants in disease prevention or control has been attributed to antioxidant properties of their constituents [14]. The most effective components seem to be flavonoids and phenolic compounds of many plant raw materials, particularly in herbs, seeds and fruits. Their metal chelating capabilities and radical scavenging properties have enabled phenolic compounds to a thought of effective free radical scavengers and inhibitors of lipid peroxidation [15].

#### ABTS scavenging activity of *Cleome viscosa* and *Trichodesma indicum*

ABTS scavenging activity of *Cleome viscosa* and *Trichodesma indicum* extracts is given in Figure 1. The Figure 1 depicts that the ethyl acetate and ethanol extracts of *Cleome viscosa* and ethanolic extracts of *Trichodesma indicum* showed the significant ( $p < 0.05$ ) ABTS scavenging activity whereas petroleum ether extracts of fruit sample of *Cleome viscosa* and *Trichodesma indicum* shows the least ABTS scavenging activity. Except that of benzene extract of leaf of *Cleome viscosa* and root of *Trichodesma indicum*, the other extracts were found to show moderate ABTS scavenging effect.

Kumar *et al.* [16] reported that ABTS and DPPH radicals, the methanolic extracts of *Amaranthus caudatus* showed significantly higher scavenging of ABTS radicals when compared to DPPH radicals. This further showed the capability of the extracts to scavenge different free radicals in different systems, indicating that they may be useful therapeutic agents for treating radical related pathological damage.

Hence the present data suggests that leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum* are capable of scavenging free radicals. Thus they may be able to prevent the initiation and propagation of free radical mediated chain reactions by stabilizing reactive species via electron or hydrogen donation before such deleterious reactions can occur.



PE- Petroleum Ether EA- Ethyl Acetate BE- Benzene ET- Ethanol CH- Chloroform

Fig. 1: ABTS scavenging activity of *Cleome viscosa* and *Trichodesma indicum*

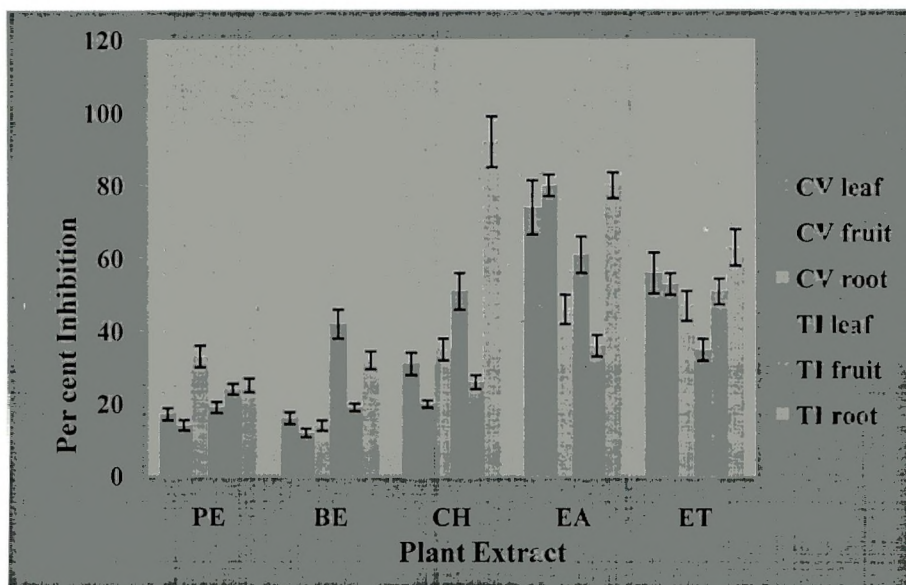
#### DPPH scavenging activity of *Cleome viscosa* and *Trichodesma indicum*

DPPH scavenging activity of *Cleome viscosa* and *Trichodesma indicum* extracts are presented in Figure 2. The figure revealed that the free

radical scavenging effect of ethyl acetate and ethanol extracts was higher than the other extracts. A significant ( $p < 0.05$ ) DPPH scavenging effect was found to be in ethyl acetate extract of leaf and fruit of *Cleome viscosa* and chloroform and ethyl acetate extracts of roots of *Trichodesma indicum*, followed by the ethanol extract of

*Cleome viscosa*. Chloroform extracts of *Trichodesma indicum* plant samples showed the good scavenging effect. The lowest level of DPPH scavenging effect was found to be in the benzene extract of fruit of *Cleome viscosa* and *Trichodesma indicum*.

Vayasthapaara Rasayana formulation was also found to be a potential DPPH-free radical scavenger since even in the concentration of 0.1 mg/ml, the activity was  $94 \pm 0.5$  per cent [17].



PE- Petroleum Ether, EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

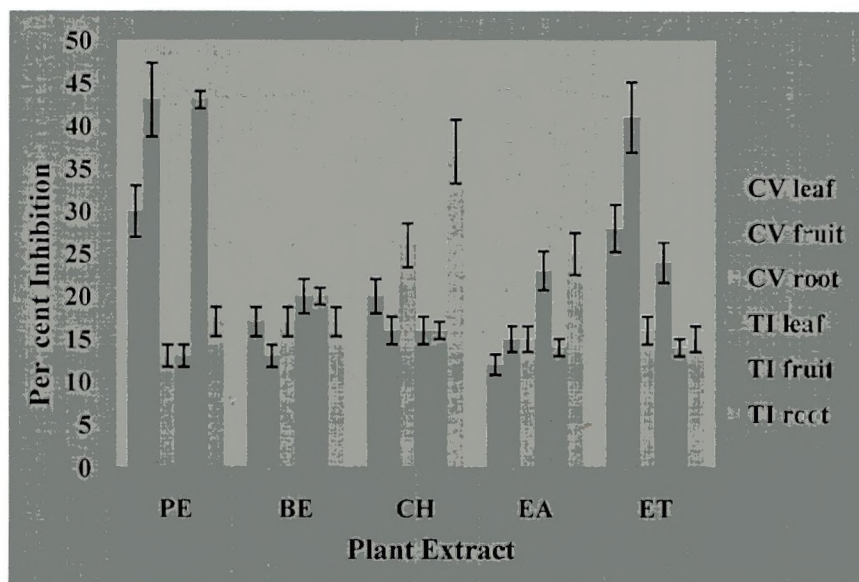
Fig. 2: DPPH scavenging activity of *Cleome viscosa* and *Trichodesma indicum*

*Clerodendrum paniculatum* shows high inhibition of DPPH activity exhibiting 54.25 per cent in leaf extract whereas 94 per cent in root extract. The results of DPPH-free radical scavenging assay suggest that the root extracts are more capable of scavenging free radicals [18].

Hence the study showed the capability of leaves, fruits and roots extracts to scavenge free radicals, indicating that they may be useful therapeutic agents for treating radical related pathological damage. Thus *Cleome viscosa* and *Trichodesma indicum* is likely to be a potential medicinal plant resources for developing antioxidants.

**Superoxide scavenging activity of *Cleome viscosa* and *Trichodesma indicum***

The results of superoxide scavenging ability of leaves, fruits and roots of *Cleome viscosa* and *Trichodesma indicum* are given in Figure 3, that the highest level of inhibitory effect of superoxide generation was observed in the petroleum ether and ethanolic extracts of *Cleome viscosa* and *Trichodesma indicum* fruit sample and the lowest inhibitory effect were shown by ethyl acetate extract of leaf of *Cleome viscosa* and petroleum ether extract of leaf of *Trichodesma indicum*. Of all the solvent extracts, the leaves and fruits samples of *Cleome viscosa* and *Trichodesma indicum* have the good inhibitory effect.



PE- Petroleum Ether, EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

Fig. 3: Superoxide scavenging activity of *Cleome viscosa* and *Trichodesma indicum*

Kaur and Arora [19], reported that the extraction of *Cassia siamea* and *Cassia javanica* leaves with different solvents showed that the ethyl acetate extract caused maximum inhibition of superoxide generation and the water extract inhibited to a lesser extent. The aqueous leaf extract of *Melastomamalabaricum*, *Dicrunopterislinearis* showed significant inhibition against superoxide generation in aqueous extracts of leaves [20].

*Cleome viscosa* and *Trichodesmaindicum* might be a good source of free radical scavengers that provide beneficial effects against oxidative damage.

#### Nitric oxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

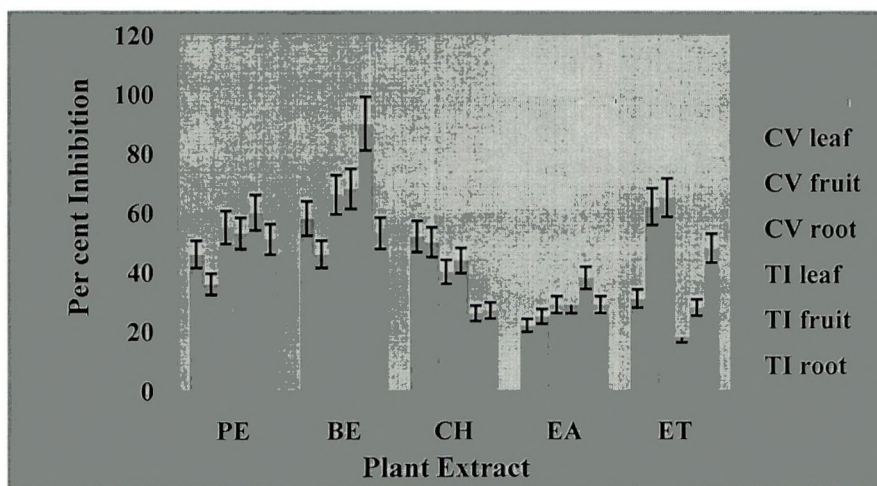
Nitric oxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum* extracts are given in Figure 4, reveals that the benzene extract of plant samples showed significant ( $p < 0.05$ ) inhibition of nitric oxide generation followed by the ethanol,

petroleum ether and chloroform extracts. Least inhibitory effect was observed in the ethyl acetate extracts of both plants. Of all the extracts fruits and roots of *Cleome viscosa* and *Trichodesmaindicum* shows the moderate activity of nitric oxide generation.

The *Clerodendronspeciosum* lowers extract had shown better nitric oxide scavenging activity than other extracts [21]. Nitric oxide (NO) is a reactive free radical generated from sodium nitroprusside in aqueous solution at physiological pH and reacts with oxygen to form nitrite [22].

Devi et al. [23] have reported that the methanolic extract of the stem (67 per cent) of *Aristolochiaindica* showed strong inhibitory action against the nitric oxide generation.

Thus the present findings indicated that *Cleome viscosa* and *Trichodesmaindicum* have very good potential against nitric oxide scavenging activity and might prevent protein and DNA damage.



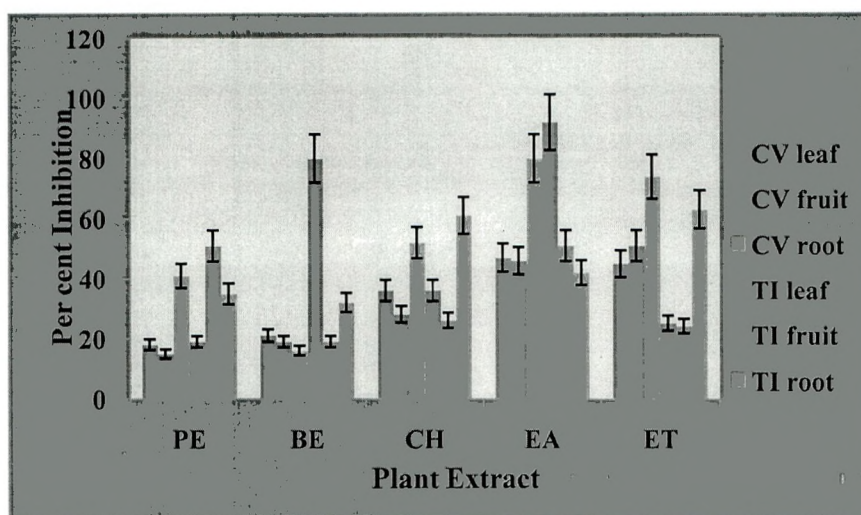
PE- Petroleum Ether, EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

Fig.4: Nitric oxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

#### Hydroxyl radical scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

Hydroxyl radical scavenging activity of *Cleome viscosa* and *Trichodesmaindicum* extracts are given in Figure 5. From the Figure 5, it is predicted that the hydroxyl ion generation was significantly

( $p < 0.05$ ) inhibited the ethyl acetate extract of root of *Cleome viscosa* and leaf of *Trichodesmaindicum*. A very low inhibition of hydroxyl radical scavenging was observed in the petroleum ether extract of fruit and benzene extract of root sample of *Cleome viscosa*. The benzene extract of leaf and chloroform and ethanol extract of root sample of *Trichodesmaindicum* shows the significant inhibitory effects.



PE- Petroleum Ether EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

Fig. 5: Hydroxyl radical scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

Maximum inhibition of hydroxyl radical generation in ethyl acetate extract of *Buteamonosperma* Lam. was documented by Lavhale and Mishra, [24].

Hence the present study showed that *Cleome viscosa* and *Trichodesmaindicum* are the potential scavenger of hydroxyl radicals. It has been suggested that the role played by *Cleome viscosa* and *Trichodesmaindicum* against hydroxyl radical more important for in lammation.

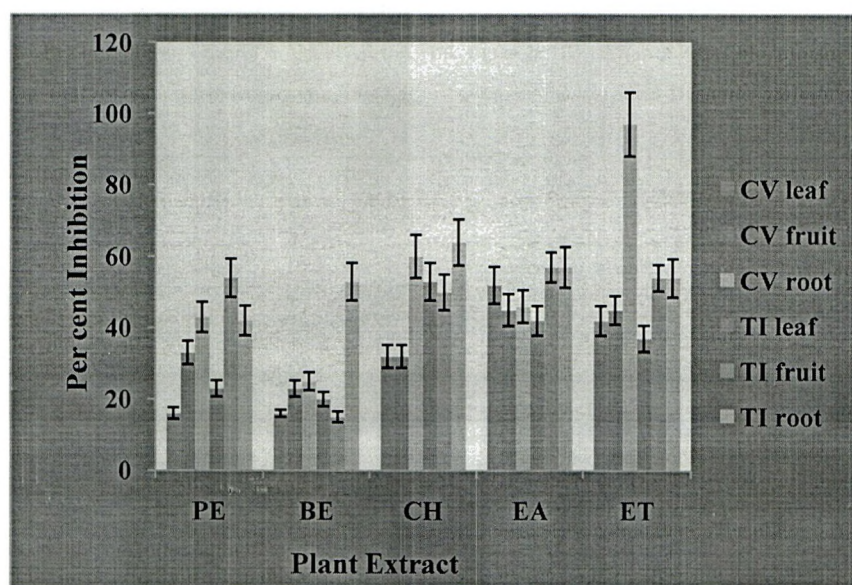
#### Hydrogen peroxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

Unsaturated lipids in cell membrane are susceptible to peroxidation. Consequently, these chain reactions initiated by hydroxyl radical attacking lipids are extended by the generated lipid hydroperoxide free radicals. Despite the low reactivity of hydrogen peroxide, the high penetrability of cellular membrane might lead to hydroxyl radical formation when it react with ferrous ion or superoxide an ion in the cell leading to oxidative stress [25].

Therefore in this study in order to consider the antioxidant property of different extracts of leaves, fruits and roots of *Cleome viscosa* and

*Trichodesmaindicum*, it is necessary to determine the scavenging activity of hydrogen peroxide by different extracts.

The results of hydrogen peroxide scavenging ability of leaves, fruits and roots of *Cleome viscosa* and *Trichodesmaindicum* are given in Figure 6, reveals that per cent inhibition of different extracts of leaves of *Cleome viscosa* showed the following order: ethyl acetate > ethanol > chloroform > petroleum ether > benzene. The per cent inhibition in different extracts of fruits of *Cleome viscosa* showed the following order: ethanol > ethyl acetate > petroleum ether > chloroform > benzene. The per cent inhibition in different extracts of roots of *Cleome viscosa* showed the following order: ethanol > chloroform > ethyl acetate > petroleum ether > benzene. The per cent inhibition in different extracts of leaves of *Trichodesmaindicum* showed the following order: chloroform > ethyl acetate > ethanol > petroleum ether > benzene. The per cent inhibition in different extracts of fruits of *Trichodesmaindicum* showed the following order: ethyl acetate > ethanol > petroleum ether > chloroform > benzene. The per cent inhibition in different extracts of roots of *Trichodesmaindicum* showed the following order: chloroform > ethyl acetate > ethanol > benzene > petroleum ether.



PE- Petroleum Ether, EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

Fig. 6: Hydrogen peroxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*

Hence this study represents the capability of the leaves, fruits and roots extracts to scavenge free radicals, indicating that they may be useful therapeutic agents for treating radical related pathological damage. Thus *Cleome viscosa* and *Trichodesmaindicum* are likely to be a potential medicinal plants resource for developing antioxidants.

The maximum activity was shown in the ethanolic extract of root of *Cleome viscosa*, whereas the chloroform and ethyl acetate extracts are showed the moderate hydrogen peroxide scavenging activity of *Cleome viscosa* and *Trichodesmaindicum*.

Hydrogen peroxide scavenging activity of the extracts of *Terminalia chebula*, *T. bellerica* and *E. officinalis* showed no substantial result compared to the standard sodium pyruvate (IC50 = 3.24) 0.30mg/ml [25].

*Ocimum sanctum* L. possess 60 per cent inhibition of hydrogen peroxide radical in ethanol extracts [26]. Hydroalcoholic extract of *Ichnocarpus frutescens* leaves exhibited 80 per cent inhibition against hydrogen peroxide scavenging activity [27].

Thus the present study showed that *Cleome viscosa* and *Trichodesmaindicum* are the potent scavengers of hydrogen

peroxide radicals. It has been suggested that the role played by *Cleome viscosa* and *Trichodesmaindicum* against hydrogen peroxide generation is more important for in lammation.

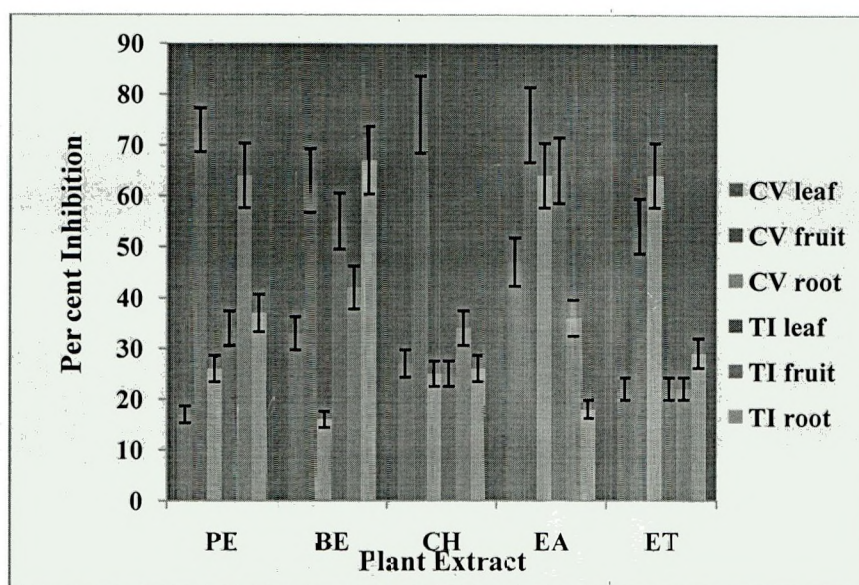
#### Inhibition of *in vitro* lipid peroxidation by *Cleome viscosa* and *Trichodesmaindicum*

Figure 7 depicts the inhibition of lipid peroxidation exerted by *Cleome viscosa* and *Trichodesmaindicum*. It is evident from Figure 7, petroleum ether, chloroform and ethyl acetate extracts of the fruits (76, 74 and 73) of *Cleome viscosa* exhibited very high inhibition. The petroleum ether extract of the leaf and benzene extract of the root of *Cleome viscosa* and ethyl acetate extract of the root of *Trichodesmaindicum* showed low inhibition than the other extracts. Among all, the fruit of *Cleome viscosa* showed significant *in vitro* lipid peroxidation than that of the other parts.

Patroet al.[29] have indicated that the ethanolic extracts of *Commiphora caudate* and *Commiphora var pubescens* scavenged the free radicals in a dose dependent manner. The water extract of *Psoralea corylifolia* L. produced the least inhibition of lipid peroxidation among chloroform, hexane and alcoholic extracts of the plant [30].

From the primary findings, it is clear that the extracts of plants *Cleome viscosa* and *Trichodesma indicum* possess antioxidant properties and could serve as free radical inhibitors or scavengers, acting possibly as primary antioxidants.

Hence to conclude the present study indicates that the leaf, fruit and root extracts of *Cleome viscosa* and *Trichodesma indicum* possessed good free radical scavenging activity and this may be used to treat the disorders caused due to free radicals.



PE- Petroleum Ether, EA- Ethyl Acetate, BE- Benzene, ET- Ethanol, CH- Chloroform

Fig. 7: Inhibition of *in vitro* lipid peroxidation by *Cleome viscosa* and *Trichodesma indicum*

## REFERENCES

- Abirami P, Gomathinayagam R, Panneerselvam O. Antioxidant activity of the medicinal plant *Enicostemma littorale* Blume, International journal of green pharmacy, 2012; 342-345.
- Mohan SC, Balamurugan V, Salini ST, Rekha R. Metal ion chelating activity and hydrogen peroxide scavenging activity of medicinal plant *Kalanchoe pinnata*, Journal of Chemical and Pharmaceutical Research, 2012; 4(1):197-202.
- Abiramasundari P, Priya V, Devi GS. Antioxidant potential and *in vitro* free radical scavenging activity of *Aervalanata*, Trop. Med. Plants, 2011; 12(1): 17-20.
- Mishra SS, Patel KK, Navdeep R, Ashish P, Prangya PP, Kundilk G, Niranjana P. Screening of ten Indian medicinal plants extracts for antioxidant activity, Ann. Biol. Res., 2011; 2(1):162-170.
- Utpal B, Vaskar B, Ghosh BK, Gunasekaran K, Rahman AA. Antinociceptive, cytotoxic and antibacterial activities of *Cleome viscosa* leaves, Rev. bras. Farmacogn. 2011; 21(1):165-169.
- Verma NV, Koche KL, Tiwari S, Mishra SK. Random amplified polymorphic DNA analysis detects variation in a micropropagated clone of *Trichodesma indicum* (L.), R. Br., Afr. J. Bio., 2010; 9(28): 4322-4325.
- Shirwaikar A, Ram HNA, Mohanpatra P. Antioxidant and antitumor activity of aqueous extracts of a polyherbal formulation, Ind. J. Exp. Biol., 2006; 44:474-480.
- Mensor LL, Menezes FS, Leitao GG, Reis AS, Dossantos T Coube CS, Ceitno SG. Screening of Brazilian plant extracts for antioxidant activity by the use of DPPH free radical method, Phytother. Res. Sci., 2001; 15: 127-130.
- Winterbourn C, Hawkins RE, Brain M, Cavell RW. The estimation of red cell superoxide dismutase activity, J. Lab. Clin. Med., 1975; 85:337-341.
- Green MJ, Hill HAO. Chemistry of dioxygen methods Enzymology, 1984; 3:105.
- Elizabeth K, Rao, MNA, Oxygen radical scavenging activity of curcumin, Int. J. Pharm., 1990; 58:237-240.
- Ruch RJ, Cheng SJ, Klauing JE. Prevention of cytotoxicity and inhibition of intra-cellular communication by antioxidant catechins isolated from Chinese green tea, J. Carcinogenesis, 1989; 10:1003-1008.
- Okhawa H, Oshicha N, Yagi K. Assay for lipid peroxide in animal tissue thiobarbituric acid reaction, Anal. Biochem., 1979; 95:351-358.
- Premanath R, Lakshmi Devi N. Studies on antioxidant activity of *Tinospora cordifolia* (Miers.), leaves using *in vitro* models, Journal of American Science, 2010; 6(10):736-743.
- Mohammad AE, Seyed MN, Seyed FN, Fatemeh B, Ahmad RB. Antioxidant and free radical scavenging activity of *H. of icinalis* L. var. *Angustifolius*, *V. Odorata*, *B. Hyrcana* and *C. Speciosum*, Pak. J. Pharm. Sci., 2010; 23(1):29-34.
- Kumar S. Free radicals and antioxidants, Human and food system, Ad. App. Sci. Res., 2011; 2(1):129-135.
- Mukherjee S, Nayana P, Omkar K, Bhagyashri N, Shrikant T, Akshay B, Pankaj P. Evaluation of free radical quenching properties of standard Ayurvedic formulation Vayasthapana Rasayana, BMC Comp. Alt. Med., 2011; (11):38.
- Arun PV, Sachin S, Suganyadevi P. Antioxidant potential of *Clerodendrum paniculatum*, Journal of Pharmacy Research, 2011; 4(6):1796-1799.
- Kaur F, Arcra S. Superoxide anion radical scavenging activity of *Cassia siamea* and *Cassia javanica*, Med. Chem. Research., 2009; 10:1-13.
- Zakaria ZA. Free radical scavenging activity of some plants available in Malaysia, Iranian J. pharmacology and therapeutics, 2007; 6:87-91.
- Ebrahimzadeh MA, Nabavi SM, Nabavi SF, Bahramian F, Bekhardnia AR. Antioxidant and free radical scavenging activity of *H. of icinalis* L. var. *Angustifolius*, *V. odorata*, *B. hyrcana* and *C. speciosum*, Pak. J. Pharm. Sci., 2010; 23(1):29-34.
- Oyedemi SO, Bradley G, Afolayan AJ. *In vitro* and *in vivo* antioxidant activities of aqueous extract of *Strychnos ningsii* Gilg, Afr. J. Pharm. Pharmacol., 2010; 4(2): 070-078.
- Devi GS, Priya V, Abiramasundari P, Archana A, Uma K, Abirami T. Antioxidant properties of the leaves and stems of *Aristolochia indica*, Plant Archives., 2010; 10(1):113-116.

24. Guo Z, Yuan Y, Ye T, Xiang J, Zhao, G. Antioxidant activities of *Salvia miltiorrhiza* and *Panax ginseng*, Food Chem., 2006; 99:767-774.
25. Lavhale MS, Mishra SH. Evaluation of free radical scavenging activity of *Buteamonosperma* Lam., Indian J. Experimental biology, 2007; 45:376-384.
26. Hazra B, Sarkar R, Biswas S, Mandal N. Comparative study of the antioxidant and reactive oxygen species scavenging properties in the extracts of the fruits of *Terminalia chebula*, *Terminalia bellerica* and *Emblica officinalis*, BMC Complementary and Alternative Medicine, 2010; 10(20):1-15.
27. Hakkim LF, Shankar GC, Girija S. Chemical composition and antioxidant property of Holy Basil (*Ocimum sanctum* L.), leaves, stems and in lorescence and their *in vitro* callus cultures, J. Agric. Food Chem., 2007; 55:9109-9117.
28. Kumarappan C, Mandal SC.  $\alpha$ -glucosidase inhibitory activity and *in vitro* antioxidant activities of alcohol-water extract of *Ischnocarpus frutescens* leaves, Med. Chem. Res., 2008; 17:219-233.
29. Patro BS, Bauri AK, Mishra S, Chattopadhyay S. Antioxidant activity of *Myristicamalabarica* extracts and their constituents, J. Agric. Food Chem, 2005; 53:6912-6918.
30. Kiran B, Raveesha KA. *In vitro* evaluation of antioxidant potentiality of seeds of *Psoralea corylifolia* L., World Appl. Sci. J., 2010; 8(8):985-990.