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Antioxidant, Antiinflammatory and Antiasthmatic Activity of *Cissus quadrangularis* Linn

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ABSTRACT

Objective: *Cissus quadrangularis* is important plant of the family *Vitaceae*. In this study stem of the plant extracted with methanol, ethanol and petroleum ether. The extract was undertaken to evaluate antioxidant, anti-inflammatory and anti-asthmatic activity of *Cissus quadrangularis* linn. Methods: The phytochemical investigation was carried out for the determination of presence of phytoconstituents. The *in vitro* antioxidant activity was carried out by using reducing power assay, hydrogen peroxide scavenging activity, DPPH scavenging activity. Anti-inflammatory activity by using *in vivo* carrageenan induced paw edema, and *in vitro* antiasthmatic activity by using isolated tracheal chain preparation. Results: All three extracts (MeCQ, EtCQ, and PECQ) showed significant antioxidant, anti-inflammatory and antiasthmatic activity. Results of our study substantiate and supports traditional and folk claim on use of *Cissus quadrangularis* for its antiasthmatic activity. Conclusion: From the data collected the mechanism of action is hypothesized to be through relieving oxidative stress, inflammation and dilation of smooth muscle.

INTRODUCTION

Asthma is a chronic inflammatory airway disease, and oxidative stress may be involved in its pathogenesis [1, 2]. Asthma affect about 300 million people worldwide and it has been estimated that further 100 million will be affected by 2025 [3]. Asthma is associated with change in the levels of eosinophils, mast cells, lymphocytes, cytokines and other inflammatory cell products. In bronchial asthma, oxidative stress aggravates airway inflammation by inducing diverse proinflammatory mediators, enhancing bronchial hyperresponsiveness, stimulating bronchospasm, and increasing mucin secretion. Although many studies have discussed the pivotal role of enhanced oxidative stress in the development and maintenance of airway inflammation, the therapeutic effects of many antioxidant agents on allergic airway inflammation are moderate at best [4].

However there is no complete remedy to cure asthma. Further several classes of synthetic drugs have been adopted in the treatment of asthma. In many conditions the patient has to be administered with drugs for a prolonged period or even lifelong. Administration of these drugs for long period may result in adverse effects / chronic toxicities or drug-drug interactions. Therefore there are several attempts to explore the possibility of natural drugs for asthma treatment [5]. Keeping this in view, a field survey was conducted to identify an herbal remedy for asthma. In the survey, we came across with *Cissus quadrangularis* which was traditionally used in asthmatic conditions by the tribals of Tamil Nadu [6]. The plant possesses activities like analgesic [7], antiinflammatory [7], osteoblastogenesis [8], restores the biomechanical properties and structure of the bone [9, 10], antitumor properties [11], reduces blood glucose levels and serum lipids [12], alleviating insulin resistance [13], gastroprotective [14], hepatoprotective [15], antiosteoporotic activity [16], antibacterial [17], antiprotozoal [18], antiplasmodial [19], antiviral activity [20], antifungal activity [21], CNS activity depressor [22], anticonvulsant properties [23]. Hence to validate the traditional use of this plant, the present study was undertaken to screen the antioxidant, anti-inflammatory and antiasthmatic activity of *Cissus quadrangularis* by using various experimental models.

MATERIALS AND METHODS

Plant material

The mature green stem of *Cissus quadrangularis* was collected locally from Nanded District, Maharashtra, India in the month of July 2011. The identification was carried out by viewing their morphological and microscopic properties and then further proceeds for authentication process. The plant herbarium was prepared, authenticated by Dr. B.D. Gachande (Associate Professor) of P.G. Department of Botany, Science College, Nanded, Maharashtra, India as (specimen S-5/NPC/2011) *Cissus quadrangularis* Linn (Family- Vitaceae). Care was taken to select healthy plant and for normal organs. From the authentication reports the drug collected was *Cissus quadrangularis* Linn Family- Vitaceae.

Preparation of the Extracts

Methanolic, ethanolic, pet. ether extracts of powdered stem were prepared in Soxhlet extractor according to the standard method till colourless solution was observed in siphon tube. 300 gm of the powdered stem and 1200 ml methanol was used for extraction. After completion of extraction solvent was cooled and dried. The extract was stored in air tight container till use. Percentage yield of extract was calculated.

Percentage yield of extract was calculated.

$$\% \text{ yield} = \frac{\text{Wt. of extract}}{\text{Wt. of sample require for extraction}}$$

Preliminary phytochemical screening [24]

Drugs and chemicals

Ethanol and petroleum ether (Fine Chem Industries, Mumbai), DPPH(1,1-diphenyl-2-picrylhydrazyl), Histamine, Acetyl choline chloride, Himedia Lab. Mumbai, Hydrogen peroxide 30% w/v, Thiobarbituric acid Loba Chem Pvt. Ltd. Mumbai.

Reducing Power Activity

The reducing power of fruit extracts was determined according to the method of Oyaizu M [25]. 2.5 ml of various concentrations of the extract, 2.5 ml phosphate buffer (0.2 M, pH 6.6) and 2.5 ml and 2.5 ml of 1% potassium ferricyanide were mixed and incubated at 50°C for 20 min and

centrifuged for 10 min at 5000 g after addition of 2.5 ml of 10% trichloroacetic acid. 2.5 ml aliquot of supernatant was mixed with 2.5 ml of deionised water and 0.5 ml of 0.1% ferric chloride. After 10 min of incubation, the absorbance was measured at 700 nm against a blank. Ascorbic acid was used as standard. The assays were carried out in triplicate and the results were expressed as Mean values ± SEM. Increased absorbance values indicate a higher reducing power.

Diphenyl-picryl-hydrazyl radical scavenging (DPPH) Assay

The scavenging effect of extracts on DPPH radicals was determined according to the method of Shimada K [26]. Various concentrations of sample (4 ml) were mixed with 1 ml of methanolic solution containing DPPH radicals, resulting in the final concentration of DPPH being 0.2 mM. The mixture was shaken vigorously and left to stand for 30 min, and the absorbance was measured at 517 nm. A reaction mixture without test sample was served as control. The percentage of inhibition can be calculated using the formula:

$$(\%)\ \text{inhibition} = \frac{A_{\text{control}} - A_{\text{test}}}{A_{\text{control}}} \times 100$$

Where,

$A_{control}$: absorbance of control

A_{sample}: absorbance of test

Hydrogen Peroxide Scavenging Activity

1ml of test extract solution [prepared in phosphate buffered saline (PBS)] with different concentrations were incubated with 0.6 ml of 4mM H_2O_2 solution (prepared in PBS) for 10 min. The absorbance of the solution was observed at 230 nm against a blank solution [27]. The concentration of H_2O_2 was spectrophotometrically intractable from absorption using the molar absorptivity of 81 M^{-1} cm^{-1}. The H_2O_2 radical scavenging activity was calculated as

$$(\%)\ \text{inhibition} = \frac{A_{\text{control}} - A_{\text{test}}}{A_{\text{control}}} \times 100$$

Where,

$A_{control}$: absorbance of control

A_{sample}: absorbance of test

***In vivo* Carrageenan induced paw edema**

Male or female Sprague-Dawley rats (150 - 200 g) kept at the laboratory Animal house was used. The animals were maintained under standard environmental conditions and had free access to standard diet and water. Anti-inflammatory activity was measured using carrageenan induced rat paw edema assay. Groups of 6 rats were given a dose of the extract (plant extracts were dissolved in sterile distilled water and administered through the P.O. route at different dose levels). After 1h, 0.1 ml, 1% carrageenan suspension in 0.9% NaCl solution was injected into the sub-plantar tissue of the right hind paw. Paw volume was measured immediately after carrageenan injection and at 30min, 1h, 2h, 3h, 4h and 5h and after 24hr by using a plethysmometer to calculate the percentage inhibition of inflammation as an index for anti-inflammatory activity [7]**.**

IAEC Approval

Male or female Sprague-Dawley rats 150 - 200 g were used for the present study. The experimental animals were maintained under standard laboratory conditions in an animal house approved by the Committee for the Purpose of Control and Supervision on Experiments on Animals (Reg. No. 1613/po/a/12/CPCSEA) under 12 h light/dark cycle and controlled temperature (24 ± 2°C) and fed with commercial pellet diet and water *ad libitum*. All animals were acclimatized to the laboratory environment for at least one week before the commencement of experiment. The experimental protocol was followed as per guidelines done by Institutional Animal Ethics Committee of Nanded Pharmacy College, Nanded (Maharashtra), India.

Animal used: Sprague-Dawley rats

Weight: 150-200 g

Sex: Male

Route of administration: P.O.

Housing Condition: Animals were housed in a group of four in separate cages under controlled conditions of temperature (24 ± 2°C). All animals were given standard diet (golden feed, New Delhi) and water regularly. Animals were further divided in eight groups with six animals in each group.

Acute toxicity studies

Acute toxicity study was carried out on methanolic, ethanolic and petroleum ether extracts of *Cissus quadrangularis* L roots on male Swiss albino mice. The mice were fasted overnight and the weight of each mouse was recorded just before use. Animals were divided randomly into ten treatment groups; each group consisting of three mice; each treatment group received orally the methanolic, ethanolic and petroleum ether extract of *Cissus quadrangularis* L roots in a dose of 5, 50, 300, 2000 and 5000 mg/kg. For each dose two groups of animals were used. Animals were kept under close observation for 4 hours after administering the extract, and then they were observed daily for three days for any change in general behaviour and/or other physical activities. Acute toxicity study was done as per OECD, 2006 Guidelines. Hence we selected 200 mg/kg and 500 mg/kg as low and high doses.

Experimental design

The following groups were made for the anti-inflammatory study. Six animals were taken for each group:

Group A: Control; Receives Carrageenan (1%)

Group B: Standard (20mg/kg); Receives 20mg/kg Diclofenac

Group C: MeCQ (200mg/kg); Receives 200mg/kg Methanolic extract of *Cissus quadrangularis* Linn

Group D: MeCQ (500mg/kg); Receives 500mg/kg Methanolic extract of *Cissus quadrangularis Linn*

Group E: EtCQ (200mg/kg); Receives 200mg/kg Ethanolic extract of *Cissus quadrangularis* Linn

Group F: EtCQ (500mg/kg); Receives 500mg/kg Ethanolic extract of *Cissus quadrangularis Linn*

Group H: Pet.ECQ (200mg/kg); Receives 200mg/kg Pet. Ether extract of *Cissus quadrangularis* Linn

Group I: Pet.ECQ (500mg/kg); Receives 500mg/kg Pet. Ether extract of *Cissus quadrangularis* Linn

EtCQ: Ethanolic extract of *Cissus quadrangularis* Linn stem

MeCQ: Methanolic extract of *Cissus quadrangularis* Linn stem

Pet.ECQ: Petroleum ether extract of *Cissus quadrangularis* Linn stem

$$\% \text{ Inhibition} = \frac{(Vt - Vo)\ \text{Control} - (Vt - Vo)\ \text{test} \times 100}{(Vt - Vo)\ \text{Control}}$$

Where: Vt: paw edema volume at time t and Vo: paw edema at predose

Antiasthmatic activity on isolated tracheal chain preparation by using kymograph paper

The goat tracheal chain was used for the experimental purpose [28]. The entire trachea was dissected out and cut into individual rings (2–3 cartilaginous rings wide). Three-four rings are tied together with threads and mounted in the organ bath containing Krebs-Henseleit solution. The tissue was maintained at 37°C under a tension of 01 g and gassed with aeration tube. Forty-five minutes are allowed for equilibration. The experiment was carried out by using students rotating drum on smoked kymograph paper. Histamine (10µg/ml) was injected via syringe, after maximum contraction had reached the test drug extracts of *Cissus quadrangularis* of varying concentrations i.e. (10mg/ml, 5mg/ml, 2.5 mg/ml) are administered separately. The tracheal responses are allowed to gradually relax at base line. Aminophyline, cyproheptidine HCl are used as standards. This experiment was similarly carried out by using acetylcholine (10ug/ml) produce contraction up to its maximum and gradual relaxation by using test drug extracts of *Cissus quadrangularis* (10mg/ml, 5mg/ ml, 2.5 mg/ml). This data was compared with standard drug atropine.

Statistical Analysis

The results were expressed as a mean ± S.E.M. The differences were compared using One Way Analysis of Variance (ANOVA) and subsequently followed by Bonferroni´s test.

RESULTS

Table 1: texture, colour and %Yield of Ethanolic, Methanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.)

Sr. no.	Drug taken(g)	Solvent used	texture	Colour of extract	Yield (gm)	%yield
1	300	methanol	Sticky	Dark brown	13.2	4.4
2	300	ethanol	Sticky	Dark brown	14.1	4.70
3	300	Petroleum ether	Non sticky	Greenish	8	2.66

Ethanolic extract gave more yield (4.70%) than methanol and petroleum ether extract while petroleum ether extracts gave least yield (2.66%).

Table 2: Phytochemical analysis of Methanolic, Ethanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.)

Chemical constituents	Methanolic Extract	Ethanolic Extract	Petroleum Ether extracts
Flavonoids	+	+	–
Carbohydrates	+	+	+
Proteins and amino acids	+	+	+
Steroids	+	+	+
Phenols and Tannins	–	+	+
Fixed oil and Fats	+	+	–
Saponins	+	+	+
Gums and Mucilage's	+	+	+
Glycosides	+	+	+

Where +: present; -: Absent

The Phytochemical qualitative screening for presence of phytoconstituents in *Cissus quadrangularis* Linn extracts reveal that- All extracts showed the presence of glycosides,

flavonoids and phytosterols and alcoholic and pet. ether extracts showed the presence of phenols and tannins.

Table 3: Antioxidant activity of Methanolic, Ethanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.) by Reducing Power Assay method

Compounds	Absorbance at 700 Nm				
	10 µg/ml	20 µg/ml	40 µg/ml	80 µg/ml	100 µg/ml
MeCQ	0.40 ± 0.006	0.52 ± 0.004	0.53 ± 0.008	0.92 ± 0.004	0.98 ± 0.003
EtCQ	0.42 ± 0.002	0.49 ± 0.008	0.54 ± 0.005	0.89 ± 0.004	0.92 ± 0.006
PECQ	0.30 ± 0.01	0.39 ± 0.008	0.49 ± 0.001	0.80 ± 0.008	0.90 ± 0.005
Std.	0.53 ± 0.007	0.67 ± 0.001	0.82 ± 0.006	0.105 ± 0.003	0.118 ± 0.004

Table 3 reveals that the methanolic extract showed 0.98±0.003 absorbance at 100µg/ml conc. while standard ascorbic acid at 100µg/ml conc. showed 0.118±0.004 absorbance measured at 700nm. It indicates more reducing power as on index of antioxidant activity.

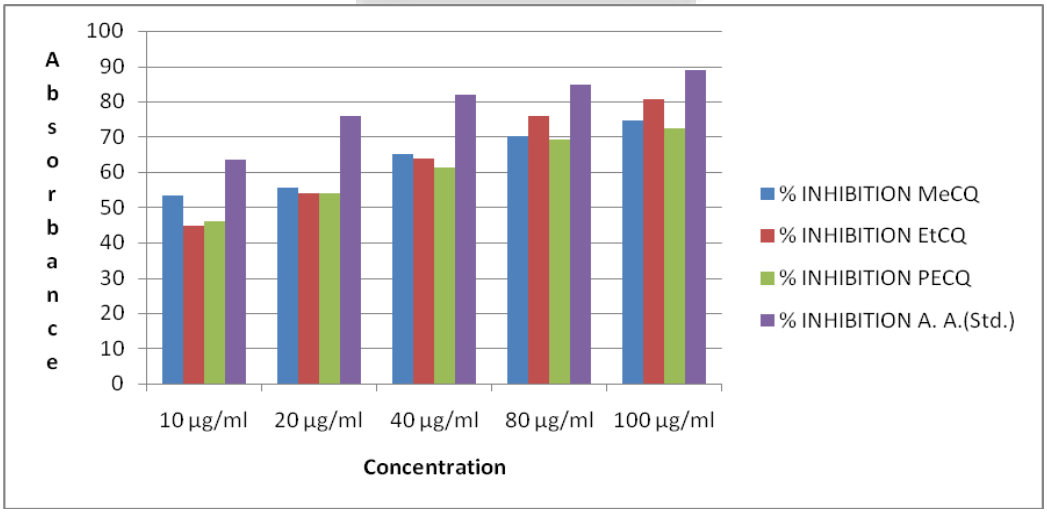

Fig. 1: Antioxidant activity of Methanolic, Ethanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.) by DPPH Method

By observing above graph it represents, the higher percentage scavenging activity of ethanolic extract **(80.58%)** than methanolic and petroleum ether extracts, as well as equivalent to the standard ascorbic acid **(88.75%).**

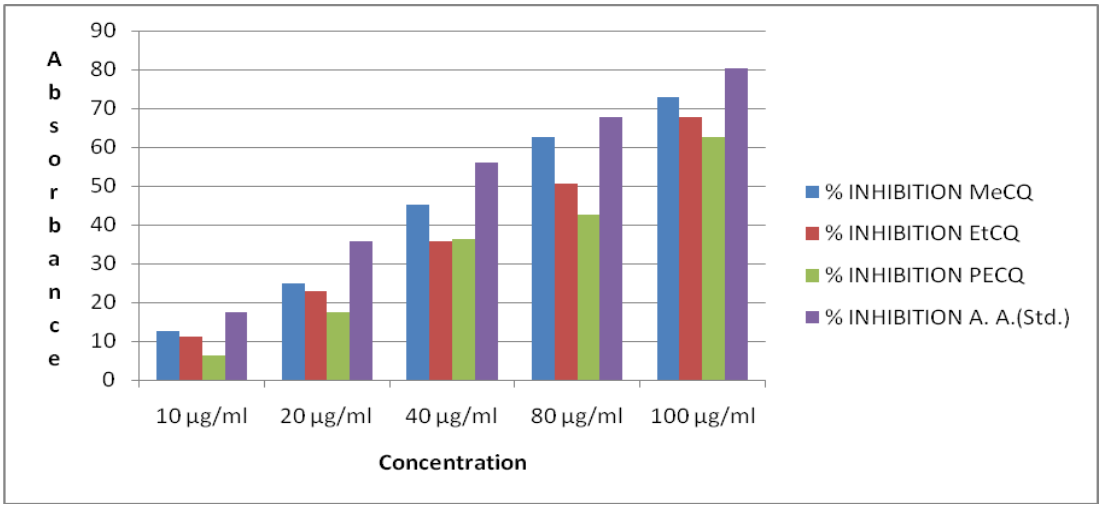

Fig. 2: Antioxidant activity of Methanolic, Ethanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.) by H_2O_2 Method

From the above figure it reveals that higher percentage scavenging activity of methanolic extract **(72.94%)** than ethanolic and petroleum ether extracts, as well as equivalent to the standard ascorbic acid **(80.16%).**

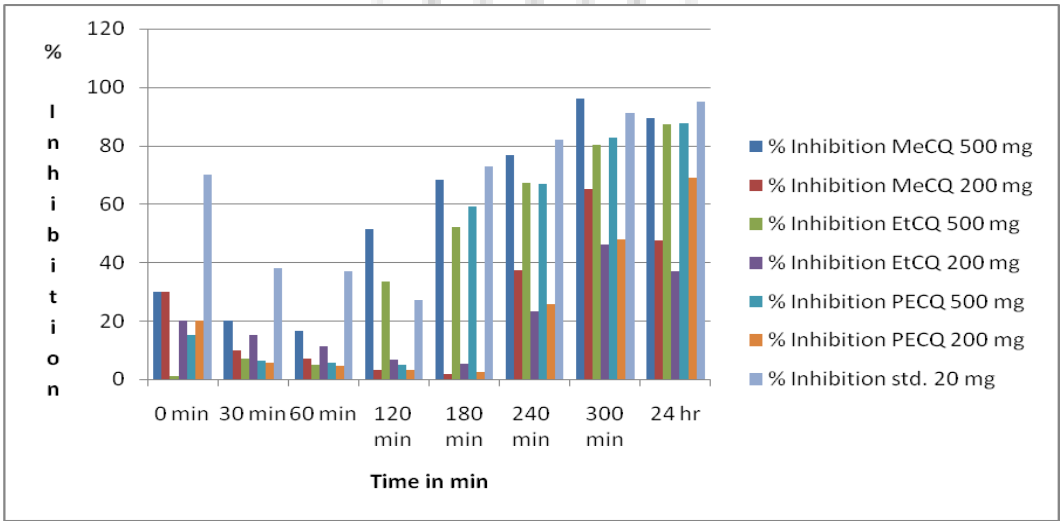

Fig. 3: Anti-inflammatory activity Methanolic, Ethanolic and Petroleum Ether extracts of *Cissus quadrangularis* (L.) by *In vivo* Carrageenan induced paw edema Method

It showed that MeCQ **(89.47%)** has potentially significant anti-inflammatory activity on comparison with EtCQ and PECQ and equivalent to Diclofenac **(95.00%)**.

Antiasthmatic activity

Table 4: Effect of MeCQ against tracheal contraction induced by Histamine (10µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation			Standard 10 µg/ml	
			2.5 mg/ml	05 mg/ml	10 mg/ml		
			Conc. of extracts			Aph.	Ch.
01	Methanolic Extract	0.2	7.69%	10%	15%	14.70%	14.28%
02		0.4	23.07%	30%	65%	29.41%	42.85%
03		0.6	38.46%	45%	100%	47.05%	61.90%
04		0.8	58.33%	65%		64.70%	80.95%
05		01	76.92%	100%		82.35%	100%
06		1.2	100%			100%	

Table 4 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml at (0.6 ml) gives 100% relaxation which is equivalent to standards Aph at (1.2 ml) and Ch at (01 ml) dose.

Table 5: Effect of EtCQ against tracheal contraction induced by Histamine (10µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation of CQ. Extract			Standard 10 µg/ml	
			2.5 mg/ml	05 mg/ml	10 mg/ml		
						Aph.	Ch.
01	Ethanolic Extract	0.2	12.00%	14.28%	16.66%	14.70%	14.28%
02		0.4	24.00%	28.57%	33.33%	29.41%	42.85%
03		0.6	36.02%	50%	50%	47.05%	61.90%
04		0.8	48.23%	64.28%	75%	64.70%	80.95%
05		01	60.00%	85.71%	100%	82.35%	100%
06		1.2	72%	100%		100%	
07		1.4	84.32%				
08		1.6	100%				

Table 5 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml at (01 ml) gives 100% relaxation which is equivalent to standards Aph at (1.2 ml) and Ch at (01 ml) dose.

Table 6: Effect of PECQ against tracheal contraction induced by Histamine (10µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation of CQ. Extract			Standard 10 µg/ml	
			2.5 mg/ml	05 mg/ml	10 mg/ml	Aph.	Ch.
01		0.2	15.38%	16.66%	19.04%	14.70%	14.28%
02		0.4	30.76%	33.33%	52.38%	29.41%	42.85%
03	Pet. ether	0.6	46.15%	66.66%	76.19%	47.05%	61.90%
04	Extract	0.8	61.15%	100%	100%	64.70%	80.95%
05		01	100%			82.35%	100%
06		1.2				100%	

Table 6 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml (0.8 ml) gives 100% which is equivalent to standards Aph (1.2 ml) and Ch (01 ml) dose.

Table 7: Effect of MeCQ against tracheal contraction induced by Acetylcholine (10µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation of CQ. Extract			Atropine 10 µg/ml
			2.5 mg/ml	05 mg/ml	10 mg/ml	
01		0.2	8.00%	11.53%	10.52%	20%
02		0.4	20.00%	23.07%	26.31%	50%
03	Methanolic	0.6	32.02%	34.61%	47.36%	70%
04	Extract	0.8	44.03%	46.51%	60%	90%
05		01	52.00%	61.53%	100%	100%
06		1.2	60.31%	76.92%		
07		1.4	88.19%	100%		
08		1.6	100%			

Table 7 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml at (01 ml) gives 100% relaxation which is equivalent to standard Atropine at (01 ml) dose.

Table 8: Effect of EtCQ against tracheal contraction induced by Acetylcholine (10 µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation of CQ. Extract			Atropine 10 µg/ml
			2.5 mg/ml	05 mg/ml	10 mg/ml	
01		0.2	05.88%	15%	10%	20%
02		0.4	17.64%	46%	25.03%	50%
03	Ethanolic	0.6	28.41%	69%	50.21%	70%
04	Extract	0.8	47.05%	100%	70.31%	90%
05		01	58.82%		90.23%	100%
06		1.2	76.00%		100%	
07		1.4	100%			

Table 8 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml at (1.2 ml) gives 100% relaxation which is equivalent to standard Atropine at (01 ml) dose.

Table 9: Effect of PECQ against tracheal contraction induced by Acetylcholine (10µg/ml)

Sr. No.	Treatment	Dose ml	% relaxation of CQ. Extract			Atropine 10 µg/ml
			2.5 mg/ml	05 mg/ml	10 mg/ml	
01		0.2	13.04%	7.69%	19.23%	20%
02		0.4	21.73%	30.76%	34.61%	50%
03	Pet. ether	0.6	34.78%	53.84%	53.84%	70%
04	Extract	0.8	47.82%	64.61%	73.07%	90%
05		01	60.35%	100%	100%	100%
06		1.2	73.91%			
07		1.4	85%			
08		1.6	100%			

Table 9 reveals all the extracts showed 100% of relaxation although dose of 10 mg/ml at (01 ml) gives 100% relaxation which is equivalent to standard Atropine at (01 ml) dose.

DISCUSSION

Three extracts were prepared with methanol, ethanol and petroleum ether. Preliminary phytochemical evaluation of all three extracts was carried out for the determination of presence of phytoconstituents. Then these extracts were used for pharmacological screening. All the extracts showed presence of phytosterols, glycosides, phenols, protein, amino acids and tannins. Presences of tricyclic triterpenoids, sreroids and other phytochemical constituents in methanolic, ethanolic and petroleum ether extracts were confirmed by Thin Layer Chromatography and Spectral Analysis. Whereas phenolic and flavonoid content was estimated by total phenolic and total flavonoid method.

Oxidative stress is commonly associated with most of the diseases and disorders, so it was thought to evaluate antioxidant property of *Cissus quadrangularis* stem. Antioxidant activity was evaluated by *in vitro* methods such as Reducing power, DPPH and Hydrogen Peroxide methods. DPPH assay is one of the most widely used methods for screening antioxidant activity of plant extracts [29]. DPPH is stable, nitrogen – centred free radical which produces violet in ethanol solution. It was reduced to a yellow coloured product, diphenyl picryl hydrazine, with the addition of the fractions in a concentration dependent manner. The reduction in the number of DPPH molecules can be correlated with the number of available hydroxyl groups. All the fractions showed significantly higher inhibition percentage (Stronger hydrogen – donating ability) and positively correlated with total phenolic content. Like the antioxidant activity, the reducing power increased with increasing the concentration. The transformation of Fe^{3+} into Fe^{2+} in the presence of various fractions was measured to determine the reducing power ability. The reducing ability of a compound generally depends on the presence of reductones. Which exert the antioxidant activity by breaking the free radical chain by donating a hydrogen atom [30]. The antioxidant principles presents in the fraction of *Cissus quadrangularis* caused the reduction of Fe^{3+}/ ferricyanide complex to the ferrous from, and thus proved the reducing power assay. In reducing power assay, the absorbance was measured which is directly proportional to antioxidant activity. Methanolic extract showed more antioxidant than ethanolic and petroleum ether extracts, while petroleum ether extract have least antioxidant activity.

In DPPH method percentage inhibition was calculated and compared with the percentage inhibition of standard (Ascorbic acid). By this method also, the ethanolic extract showed highest

percentage of inhibition at 100µg/ml was 80.58%. By all these antioxidant methods it is proved that *Cissus quadrangularis* Linn stem has a good antioxidant activity and may be used in many diseases occurred due to oxidative stress such as asthma, cancer, pulmonary oedema etc. In Hydrogen Peroxide Scavenging Activity [31] percentage inhibition was calculated and compared with the percentage inhibition of standard (Ascorbic acid). Methanolic extract showed more antioxidant than ethanolic and petroleum ether extracts. Methanolic extract at 100µg/ml showed 72.94 % of inhibition where ascorbic acid showed 80.16 % of inhibition.

Carrageenan induced rat paw oedema model is widely used animal model used for evaluation of novel anti-inflammatory compounds [32]. Inflammation induced by carrageenan is biphasic response. The early phase of the inflammation is due to the release of histamine, serotonin and similar substances; and the later phase is associated with the activation of kinin-like substances, i.e., prostaglandins, proteases and lysosome [32, 33]. The previous studies have reported that bronchial asthma is common clinically diverse condition with an appreciable infiltration of inflammatory components. Hence, *in vivo* anti-inflammatory activity was carried out by using Carrageenan induced paw edema. In this method rats were used for activity and the paw edema volume was measured by using digital plethysmometer. Diclofenac was referred as standard. MeCQ extract reveals better anti-inflammatory activity than EtCQ and PECQ extracts and equivalents to standard diclofenac drug**.**

Bronchial asthma is commonly characterized by increased airway reactivity to spasmogens. An initial event in asthma appears to be the release of inflammatory mediators like histamine, triggered by exposure to allergens that directly cause acute bronchoconstriction [34, 35]. Increasing evidence suggests that the frequently observed association between activated T lymphocytes and eosinophils plays a major role in the development of airway inflammation and in the accompanying bronchial hyper-reactivity. Neutrophils and monocytes play a pivotal role in the disease process as they are a source of variety of inflammatory mediators which are responsible for bronchial hyper-responsiveness and airway inflammation [37].

In this study *Cissus quadrangularis* Linn showed potent antiasthmatic activity supporting its traditional use. The property was established by *in vitro* antiasthmatic activity on isolated tracheal chain preparation, method used maximum contraction by agonist followed by gradual relaxation by using 2.5mg/ml, 5mg/ml and 10mg/ml concentrations of three (MeCQ, EtCQ,

PECQ) extracts. The agonists were used are histamine and Ach. The activity was conducted on kymograph paper. The study was compared by using standard aminophyline, cyproheptidine against histamine contractile tissue and atropine against acetylcholine contractile chain.

The data was represented by percentage relaxation of tracheal muscle chain. The all extracts showed 100% relaxation in sequence of 2.5mg/ml < 05mg/ml < 10 mg/ml i.e. 10mg/ml conc. were much more potential of all extracts and also equivalent to the standard drugs.

CONCLUSION

The results of the present study demonstrated that the qualitative analysis of methanolic, ethanolic and petroleum ether extracts of *Cissus quadrangularis* Linn confirmed the presence of flavanoids, tannins, phenols, carbohydrates, glycosides, saponin, steroids, gums and mucilage. The antioxidant, anti-inflammatory and antiasthmatic activity of *Cissus quadrangularis* Linn were also confirmed. All three studies are correlated with each other to evaluate antiasthmatic activity because asthmatic condition occurred with oxidative stress with inflammatory symptoms.

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