CENTRAL NERVOUS SYSTEM DEPRESSANT ACTIVITY OF AQUEOUS EXTRACT OF *CISSUS QUADRANGULARIS*

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ABSTRACT

The research work deals with the screening of aqueous extracts of *Cissus quadrangularis* (CQ) stem for central nervous system (CNS) depressant activity. The CQ leaves and stems contain resveratrol, piceatannol, pallidol perthenocissin derivatives as chemical components and are reported to have anticonvulant activity. The extract of plant showed $GABA_A$-benzodiazepine receptor binding activity we made an attempt to study its CNS depressant effect. The different activities studied were test for locomotor activity, effect on muscle co-ordination, antiaggressive and antianxiety activities. The result of the study reflected that aqueous extract of the stem (150 mg/kg, p.o) decreased locomotor activity, produced muscle relaxation and showed antianxiety and antiaggressive activity

KEY WORDS: Antianxiety, Antiaggressive ,*Cissus quadrangularis*, Muscle coordination.

INTRODUCTION

Cissus quadrangularis Linn. a climber in the family Vitaceae, is one of the most frequently used medicinal plants in India. Pharmacological studies revealed the bone fracture healing property, this plant reported the antibacterial and antioxidant activities of the extract from *Cissus quadrangularis* (CQ) commonly known as 'bone setter', is frequently used as a common food item in India. The extract of plant showed $GABA_A$- benzodiazepine receptor binding activity[1]. Its anti-inflammatory effect could be produced by the flavonoids especially luteolin, and by _-sitosterol. The venotonic effect of CQ may also be postulated to be due to the effect of flavonoids present in the extract which act in the same way as that of diosmin and hesperidin. As diosmin and hesperidin are used in combination to treat hemorrhoid, the extract which produced the same activities (anti-inflammatory and

venotonic) can also be used as antihemorrhoidal drug[2]. The antiosteoporotic activity of CQ maybe justifiably is attributed to the steroids present which probably act as phyto-estrogens to effectively prevent or reduce bone loss[3]. Ethanlic extract of CQ inhibits gastric damage by enhancement in antioxidant defence enzymes in gastric mucosal tissues. In addition, ulcer protection was confirmed by histoarchitecture, which was comprised of reduced size of ulcer crater and restoration of mucosal epithelium[4], produced significant immunomodulatory effect, antioxidant activity along with the ability to modulate PG synthesis and up-regulation of the growth factors[5]. Besides these effects, the present study showed that CQ also possesses analgesic effect, which can be very useful in painful hemorrhoid. The aqueous extract of CQ was reported to have anticonvulsant and sedative property[6]. Phytochemical studies of CQ found several phytochemical constituents such as flavonoids, triterpenoids, stilbene derivatives and many others, e.g. resveratrol, piceatannol, pallidol perthenocissin, and phytosterols.

MATERIAL AND METHODS

EXTRACT PREPARATION: Fresh stems of *Cissus quadrangularis* were collected and authenticated by Mr. Sanjay Nandanwar, from an area in Punjabrao Krushi Vidyapeeth, Akola. (MH) India. The stems were washed and cut in to pieces and air dried. The powder (250 g) was macerated for 3 days in 2.5 l of distilled water at room temperature. The mixture was filtrated with a Watman n°1 filter paper and the filtrate was evaporated using a Rota vapor at a temperature of 70°C. The quantity of aqueous extract of *Cissus quadrangularis* (ACQ) obtained after evaporation was 16 g that represent a 6.4% yield, before use, the extract was dissolve in distilled water for administration orally.

ANIMALS: The male Spragua dwaley rats weighing 180 ± 5 g were selected for this study [Approved by the institution animal ethical committee (Reg.No.KMCP/08/3-23)]. The rats were housed in clean polypropylene cages having 6 rats per cage and maintained under temperature controlled room (27±2° C) with photoperiod of 12h light and 12h dark cycle. The animals were fed with commercially available food pellet diet and water *ad libitum.*

DRUGS AND CHEMICALS: Diazepam (Ranbaxy laboratories ltd., Mumbai) was used as a standard anxiolytic agent. The other entire chemicals were of analytical grade.

EXPERIMENTAL METHOD

ELEVATED PLUS-MAZE TEST[6,7] (EPM): The EMP consisted of two open arms (35×5 cm) crossed with two closed arms (35×5×20 cm). The arms were connected together with a central square of 5×5 cm. the apparatus was elevated to the height of 25 cm in a dimly illuminated room. Rats were treated with ACQ (50, 100, and 150 mg/kg) [6,7], diazepam (1 mg/kg, i.p), or vehicle 30 min before being placed individually in the centre of the EMP, facing a closed arms was recorded for 5 min. The numbers showed significant increase in exploratory activity ($P < 0.01$).

OPEN FIELD TEST[8,9,10]: The apparatus consisted of wooden box (60×60×30 cm).

The floor of the box was divided into 16 squares (15×15 cm). The apparatus was illuminated with a 40-W lamp suspended 100 cm above. Rats were treated with ACQ (50, 100 and 150 mg/kg) [8,9,10] or vehicle. After 30 min, they were placed individually in one of the corner squares; the numbers of squares crossed were counted or 5 min. Diazepam (1 mg/kg, i.p.), was used as the positive control drug.

LOCOMOTOR ACTIVITY[7,8] : The locomotor activity was measured using an actophotometer. The movement of the animal interrupts a beam of light falling on a photocell, at which a count was recorded and displayed digitally. Each rat was placed individually in the actophotometer for 10 min and the basal activity score was obtained. Subsequently, the animals were divided into groups, each consisting of six animals. ACQ (50, 100, and 159 mg/kg) [8,9,10], vehicle, or diazepam (1 mg/kg, i.p) was administered and after 30 min the mice were placed again in the actophotometer for recording the activity score.

STATISTICAL ANALYSIS

The statistical analysis was performed using the one- way ANOVA followed by students Newman Keul's Multiple range test. Results are expressed as mean ± S.E.M.

RESULTS AND DISCUSSION

Results indicate that ACQ (100 and 150 mg/kg) and diazepam (1 mg/kg) induce significant ($P < 0.01$) increase in the occupancy in the open arm. However, ACQ in the dose of 50 and 100 mg/kg did not cause a significant decrease in the time spent in the closed arm (Table 1). In open field test ACQ and diazepam brought about a significant ($P < 0.01$) and dose dependant increase in the number of squares transverse (Table 2). ACQ in doses of 100 and 150 mg/kg produced significant ($P < 0.01$) reduction in locomotor activity as compared to the control animals (Table 3).

On considering most anxiety disorders their etiology is not yet fully understood, but the picture becoming beat of clear in recent past. As it is well known Benzodiazepines (BZDs) are durg of choice and relatively safe widely used anxiolytic agents. In anxiety role of GABA is well established. Undoubtly, these agents are known to act through the BZD-GABA receptors. Despite the widespread traditional use of *Cissus quadrangularis* for treating various disorders no reports on its pharmacological effects. The present work demonstrated that the aqueous extract of CQ had anti anxiety, locomotor activity, effect on muscle co-ordination and antiaggressive effect in several behavioral parameters, like elevated pluse maze, open field and light/dark paradigms. The EMP is one of most popular animal test for researcher on behavioral pharmacology of anxiety. In EMP, animal will normally prefer to spend much of their allotted time in the closed arms. This preference reflects an aversion towards open arms that is generated by fear of open spaces. Drugs that increases open arms exploration are considered as anxiolytic effect. In our study, we observed that ACQ (100 and 150mg/kg) induced significant increases in the both the number of entries and time spent in open arms. The number of entries and the time spent in closed arms were

reduced in the extract treated group as compared to the control group. The result obtained in the open field test showed that ACQ administration significantly increases rearing, assisted rearing, and number of squares transverse, which supports the anxiolytic like activity of ACQ.

The light and dark paradigm is based on thenatural aversion of anio brightly lit places. Anxiolytic reduce the natural aversion to light ad increase the time spent in lit compartment. In this model, compared to vehicle, ACQ produced significant increase in the time spent in lighted box and decrease in the tme spent in the dark box, thus showing its anxiolytic activity.

Locomotor activity, an index of alertness and its decrease indicates sedative-hypnotics action of the BZDs make them most the most preferable and important $GABA_A$-modulating drugs. The mechanism of anxiolytic action of ACQ might involve an action on GABAergic mechanism of transmission. However, as crude extract administered for screening of CNS depressant activity of ACQ, further studies are needed to ascertain these activities of active constituent of CQ solely responsible for this.

CONCLUSION

Concluding from earlier chemical constituent reported on plant CQ having several phytochemical such as flavonoids, triterpenoids, stilbene derivatives and many others, e.g. resveratrol, piceatannol, pallidol perthenocissin, and phytosterols. The results obtained in this study suggest that the extract of the stems of *Cissus quadrangularis* possesses anxiolytic effect, locomotor activity, and effect on muscle co-ordination, antiaggressive effect in rats in several behavioral parameters, like elevated plus-maze, open field, and locomotor activity which is possibly mediated through the $GABA_A$-BZD mechanism. This study provides further scientific evidence of antideperssent activity of aqueous extract of CQ stems on oral admistration of crude extract. It further proposed that at it shows anxiolytic and antiaggressive effect it contributes to possess sedative and anticonvulsant properties.

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REFERENCES

1. Ayyanar M and Ignacimuthu S. Pharmacological actions of Cassia auriculata L. and Cissus quadranularis Wall.: A Short Review. Journal of Pharmacology and Toxicology 2008, (3): 213-221.
2. Panthong A, Supraditaporn W, Kanjanapothi D, Taesotikul T, Reutrakul V. Analgesic, anti-inflammatory and venotonic effects of *Cissus quadrangularis* Linn., Journal of Ethnopharmacology 2007, (110): 264–270.
3. Shirwaikar A, Khan S, Malini S. Antiosteoporotic effect of ethanol extract of *Cissus quadrangularis* Linn. on ovariectomized rat, Journal of

Ethnopharmacology, 2003, (89): 245–250.
4. Jainu M, Srinivasulu C, Devi S. Gastroprotective action of *Cissus quadrangularis* extract against NSAID induced gastric ulcer: Role of proinflammatory cytokines and oxidative damage, Chemico-Biological Interactions, 2006, (161): 262–270.
5. Jainu M, Mohan KV. Protective role of ascorbic acid isolated from *Cissus quadrangularis* on NSAID induced toxicity through immunomodulating response and growth factors expression, International Immunopharmacology, 2008, (8):1721–1727.
6. Bum E N, Ngoupaye GT, Talla E, Dimo T, Nkantchoua GCN, et al. The anticonvulsant and sedative properties of stems of *Cissus quadrangularis* in mice, African Journal of Pharmacy and Pharmacology, 2008, (2): 042-047.
7. Cruz-Morales SE, Santos NR, Brandao ML. One-trial tolerance to midazolam is due to enhancement of fear and reduction of anxiolytic-sensitive behaviors in the elevated plus-maze retest in the rat, Pharmacology, Biochemistry and Behavior, 2002, (72): 973–978.
8. Torres C, Escarabajal MD. Validation of a behavioral recording automated system in the elevated plus-maze test, Life Sciences, 2002, (7): 1751–1762.
9. Kumar A, Padmanabhan N and Krishnana MRV. Central Nervous System Activity of Syzygium cumini seed, Pakistan Journal of Nutrition, 2007, 6, 698-700.
10. Rex A, Voigt J P, Voits M and Fink H. Pharmacological Evaluation of a Modified Open-Field Test Sensitive to Anxiolytic Drugs, Pharmacology Biochemistry and Behavior, 1998, 59: 677–683.

Table1: Effect of ACQ on animals stay in the open and closed arms of the elevated plus maze

Treatment	Time spent in open arm(s)	Time spent in enclosed arm(s)	Entries in to open arm	Entries in to closed arm
Vehicle	30.8±6.4	243.5±8.8	8.3±1.26	14.7±1.0
Diazepam	107.0±12**	169.0±13.1**	20.5±1.9**	9.7±0.9**
ACQ(50 mg/kg, oral)	36.50±9.3	218.7±8.5	7.3±1.4	11.5±1.3
ACQ(100 mg/kg, oral)	89.8 ± 8.9	216.8±9.6	9.2±1.3	10.7±1.4
ACQ(150 mg/kg, oral)	117.0±7.1**	177.2±8.6**	17.5±1.7**	10.5±1.0

N=6 for open arm, N=6 for closed arms, **P<0.01 Vs vehicle
(One way ANOVA followed by students Newman Keul's Multiple Range test)

Table 2: Effect of ACQ on rearing and locomotion in the open field model

Treatment	Rearing	Assisted rearing	No. of square traversed
Vehicle	4.0±1.0	11.5±1.2	89±4.7
Diazepam	16.0±1.5**	21.7±3.3**	169.5±7.9**
ACQ(50 mg/kg, oral)	12.0±1.2	13.2±1.2	125.2±7.5**
ACQ(100 mg/kg, oral)	22.5± 4.5**	20.5±1.8*	140.5±8.2**
ACQ(150 mg/kg, oral)	30.3±2.5**	20.8±2.0*	158.7±8.1**

N=6 *P<0.05, **P<0.01 Vs vehicle
(One way ANOVA followed by students Newman Keul's Multiple Range test)

Table 3: Effect of ACQ on a locomotor activity in rats, assessed using the actophotometer

Treatment	Locomotor activity (score) in 10 min		% Change in activity
	Before treatment	After treatment	
Vehicle	207.52±19.50	201.0±9.44	-
Diazepam	187.50±20.09**	90.30±9.77**	55.07**
ACQ(50 mg/kg, oral)	230.30±23.59	97.16±16.55	67.41
ACQ(100 mg/kg, oral)	217.32±30.80**	141.0±33.89	29.85
ACQ(150 mg/kg, oral)	214.83±22.48**	39.67±14.6**	84.27

N=6 **P<0.01 Vs vehicle
(One way ANOVA followed by students Newman Keul's Multiple Range test)