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EVALUATION OF ANTIDEPRESSANT ACTIVITY OF AQUEOUS EXTRACT OF *BRASSICA OLERACEAE* VAR. *CAPITATA* F. *RUBRA* IN MICE

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ABSTRACT
Objective: To evaluate the antidepressant activity of aqueous extract of *Brassica oleraceae* var.*capitata* f. *rubra*.(AEBOVCR). **Methods:** Swiss albino mice were treated with two different doses of the extracts (i.e. 250 and 500 mg/kg orally) and behaviour was observed on the FST, TST, HBT. The immobility time was evaluated for the FST, TST and number of head dips for the HBT .The evaluation was done on the 0th day (before treatment) and on the 11th day (after treatment). **Results**: The extract showed significant dose dependant antidepressant activity in terms of decrease in immobility time in both FST and TST and increase in number of head dips in HBT. **Conclusion**: The findings of the study suggest that the AEBOVCR posses antidepressant like property in mice.

KEYWORDS: *Brassica oleraceae* var.*capitata* f. *rubra*, red cabbage, antidepressant, forced swimming test, tail suspension test, holeboard test.

INTRODUCTION
Depression is one of the top five most prevalent diseases worldwide. By 2020, it is expected to be the second-leading cause of disability globally. Various pharmacological agents like selective serotonin inhibitor, tricyclic antidepressant, mono amino oxidases are available. Despite the advances in pharmacotherapy about significant proportion of patients with depression are considered treatment resistant and are accompanied with number of adverse reactions.[1] In the search for new therapeutic products for the treatment of neurological disorders, medicinal plant research, worldwide, has progressed constantly, demonstrating the pharmacological effectiveness of different plant species in a variety of animal models.[2]

Brassica oleraceae var.*capitata* f. *rubra* (BOVCR**)** a member of Brassicaceae family is polyphenolic rich vegetable proven for its excellent antioxidant capacity. Due to its bioactive constituents it has several health promoting benefits.[3-6] Literature mentions BOVCR in the Phytochemical and Ethnobotantical Databases list of antidepressant.[7] However there is no scientific data to prove the claim. Furthermore depression and anxiety are also correlated with a lowered total antioxidant state. The classical antidepressants may produce therapeutic effects other than regulation of monoamines by increasing the antioxidant levels. Literature review revealed that polyphenols modulate monoaminergic neurotransmission in the brain and thus possess antidepressant-like activity at least in animal models of depression.[8] Hence the present study was carried out.

MATERIALS AND METHODS
BOVCR was collected from a local market in Mangalore, India and was authenticated by a botanist of St. Aloysius College, Mangalore.

Preparation of the extract
The leaves of BOVCR were shade dried, blended in the mixer. It was then extracted by Cold Maceration method. 100 gm of powdered drug in 200 ml of water in conical flask sealed by aluminium foil was cold macerated for 3 days on a mechanical shaker. The supernatant obtained was evaporated with rotary flash evaporator to obtain semisolid residue and stored in a refrigerator.

Phytochemical screening
The AEBOVCR was subjected to standard phytochemical screening tests for various phytoconstituents.

Experimental animals
Swiss albino mice (22-25g) of either sex were procured from animal house of Srinivas College Of Pharmacy, Mangalore. They were maintained under standard

conditions (temperature 22 ± 2oC, relative humidity 50±5% and 12 h light/dark cycle). The animals were housed in sanitized polypropylene cages containing sterile paddy husk as bedding. They had free access to standard pellet diet and water. The Institutional Animal Ethics Committee approved the experimental protocol (approval no SCP/IAEC/F150/P116/2017). All the animals received human care according to the criteria outlined in the "Guide for the Care and Use of Laboratory Animals" prepared by the "National Academy of Sciences" and published by the "National Institute of Health". Animals were acclimatized for atleast one week before use.

Treatment

Wister mice (20 - 25g) of either sex were randomly divided into four groups of six animals each. The different groups are as below.

Group I : normal control
Group II: Standard group Imipramine, (10 mg/kg,) p.o.
Group III: Test group AEBOCR (250mg/kg, p.o.)
Group IV: Test group AEBOCR (500mg/kg, p.o.)

Forced swimming test (FST)[9]

Mice was forced to swim individually for 15 min, in a glass beaker of 11cm diameter, 15cm height containing fresh water up to a height of 6cm, at a temperature of 27±2^{o}c. This constituted the "pre-test" session. The test-session is conducted on (0th day) and after the drug treatment (on 11th day). The mouse was considered immobile when it floats motionlessly or makes only those movements necessary to keep its head above the water surface. The total duration of the immobility during the 6 min test will be recorded.

Tail suspension test (TST)

Mice was suspended by tail from a height of 75cm .The mouse makes attempts to regain upright posture, but continues in a motion less state (immobility phase). Baseline immobility was measured for a period of 6 min". The test-session will be conducted on (0th day) and after the drug treatment (on 11th day) immobility will be recorded for a period of 6min.

Hole board test (HBT)[10]

Drugs were administered for 11 days. Hole board consists of a wooden board having dimensions of 40 × 40 × 25 cm^{3} along with uniformly dispersed 16 holes each having constant diameter of 3 cm. Animal was placed at the centre of hole board before and after the administration of given treatments and its activity is observed for a duration of 5 min. The number of head dips were taken into account in this behavioural paradigm.

Statistical analysis

All data were expressed as Mean±SEM. The statistical significance between groups were compared using o ANOVA, followed by Turkey (multiple comparison test).

RESULTS

Preliminary pytochemical screening

Phytochemical screening revealed the presence of flavanoids, glycosides, carbohydrates, tannins, saponins.

Table 1: Effect of AEBOVCR on duration of immobility in forced swim test.

Group	dose	No. of animals	Duration of immobility (seconds)	
			Before treatment	After treatment
control		6	148.7±5.886	147.2±5.521
Imipramine	10mg/kg	6	146.08±5.756	70.50±3.442***
AEBOVCR (low dose)	250mg/kg	6	147.05±6.126	116.5±6.725*
AEBOVCR (high dose)	500mg/kg	6	146.29±6.041	110.20±4.223**
Values are expressed as the mean ± SEM. n=6. Data were analyzed by one-way ANOVA followed by Tukey's comparision test. *p<0.05, **p<0.01, ***p<0.001, as compared to depressive control mice.				

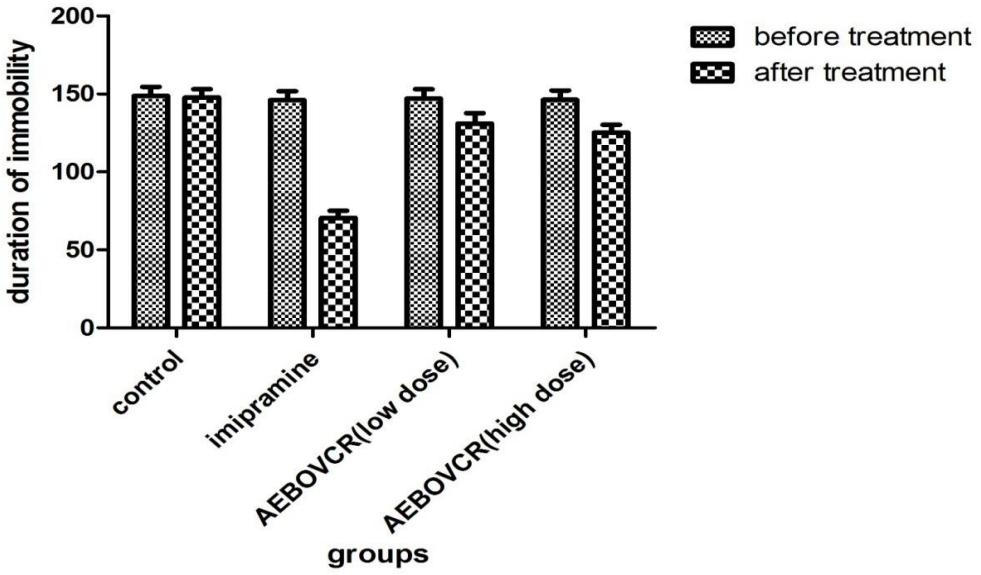

Fig 1: Effect of AEBOVCR on duration of immobility in forced swim test.

Table 2: Effect of AEB0VCR on duration of immobility in tail suspension test.

Group	dose	No. of animals	Duration of immobility (in seconds)	
			Before treatment	After treatment
control		6	139.3±5.287	136.0±4.826
Imipramine	10mg/kg	6	140.8±4.844	73.30±5.477***
AEBOVCR (low dose)	250mg/kg	6	141.0±6.136	131.50±6.326*
AEBOVCR (high dose)	500mg/kg	6	141.2±5.418	123.80±4.637**
Values are expressed as the mean ± SEM. n=6. Data were analyzed by one-way ANOVA followed by Tukey's comparision test. *p<0.05, **p<0.01, ***p<0.001, as compared to depressive control mice.				

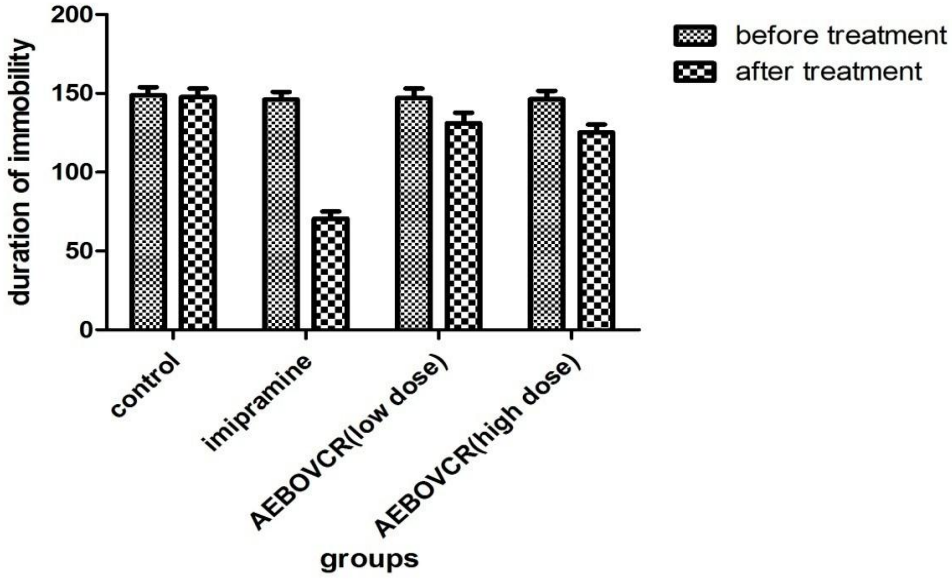

Fig 2: Effect of AEB0VCR on duration of immobility in tail suspension test.

Table 3: Effect of AEBOVCR on number of head dips in hole board test.

Group	Dose	No. of animals	Number of head dips	
			Before treatment	After treatment
control		6	68.83± 5.102	70.67±3.430
Imipramine	10	6	69.67± 6.528	123.30±5.120***
AEBOVCR (low dose)	250 mg/kg	6	69.00± 6.215	90.00±5.832*
AEBOCR (high dose)	500 mg/kg	6	68.00±3.127	98.11±5.250**
Values are expressed as the mean ± SEM. n=6. Data were analyzed by one-way ANOVA followed by Tukey's comparision test. *p<0.05, **p<0.01, ***p<0.001, as compared to depressive control mice.				

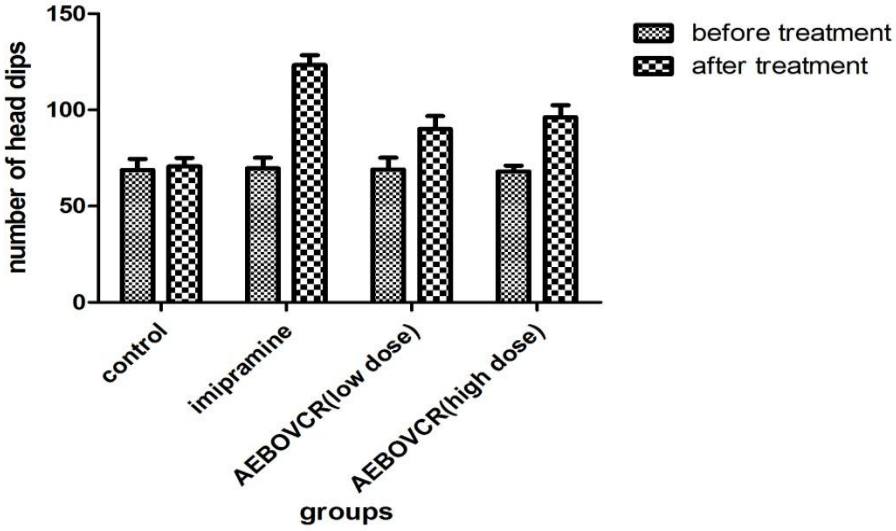

Fig 3: **Effect of AEBOVCR on number of head dips in hole board test.**

DISCUSSION

Acute stress is used in animal models to induce behavioural, physiological and neural changes relative to human depression.[11] The TST and FST are two validated models used to assess putative antidepressants compounds. Immobility time in these two paradigms reflects antidepressant-like activity. The development of immobility when rodents are suspended by their tail during TST and when they are placed in an inescapable cylinder of water during FST reflects the cessation of their persistent escape-directed behavior. Conventional drugs reliably decrease the duration of immobility in animals during these tests. This decrease in duration of immobility is considered to have a good predictive value in the evaluation of potential antidepressant agents.[12]

There is growing evidence that the imbalance between oxidative stress and the antioxidant defense system may be associated with the development neuropsychiatric disorders, such as depression and anxiety. Major depression and anxiety are also correlated with a lowered total antioxidant state and by an activated oxidative stress (OS) pathway. The classical antidepressants may produce therapeutic effects other than regulation of monoamines by increasing the antioxidant levels and normalizing the damage caused by OS processes.[13] It has been found that natural polyphenols are active in these animal models of despair. Neurochemical estimation has shown that some of these polyphenols modulate various neurotransmitters in the brain that are involved in the pathophysiology of various neuropsychiatric and neurodegenerative disorders.[8]

BOVCR is one of the richest source of antioxidants which is enriched with phenolic compounds. Several authors studied the polyphenolic profile and the antioxidant capacity of BOVCR. The strongest antioxidant capacity of BOVCR is attributed to anthocyanins which has a power of 150 flavanoids.[14]

In the present study antidepressant activity of AEBOVCR was confirmed in terms of decrease in immobility time in both FST and TST models and increase in number of head dips in HBT. AEBOVCR significantly produced dose dependant antidepressant action in the experimental models. The exact mechanism of action however is unknown but it may be endorsed to the rich antioxidant capacity by the individual or combined action of phyto constituents present in BOVCR.

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