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Anti-stress activity of different compositions of Panchagavya and *Aloe Barbedansis* Mill by using Force Swimming Method

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ABSTRACT

Panchagavya is an incredible source for many medicinal substances. It has been reported for synergistic action but their scientific data are not available. Sixty mice were randomly divided into ten groups. The first, second, third, fourth, fifth, sixth, seventh, eighth, ninth & tenth groups received PG 1, PG1+ EEAB 10%, PG1+ EEAB 50%, PG1+ EEAB 75%, PG2, PG2+ EEAB 10%, PG2+ EEAB 50%, PG2+ EEAB 75%, Standard Alprazolam, Control Urine every day administered at the dose 4ml/kg body weight regularly at 9:00 am for 21 days & investigated the role of different composition of Panchagavya and its ethanolic extract of Aloe barbedansis Mill (EEAB) (Xanthorrhoeaceae) for synergistic Anti-stress activity by using Forced Swimming Method in Swiss albino mice. On the 1st, 6th, 11th, 16th & 21th day after drug administration, effect of PG 1, PG 2, PG 1 + EEAB and PG 2 + EEAB were found to be significant at the level $p < 0.01$ as compared to Standard and Control group. The synergistic activity of PG with EEAB might be due to enhanced brain Gamma Amino Butyric Acid (GABA) level of mice and decreased brain dopamine levels & plasma corticosterone levels.

Key words: Ethanolic extract of *Aloe barbedansis* Mill (EEAB), Anti-stress activity, Panchagavya

INTRODUCTION

In psychology, stress is a feeling of strain and pressure. Symptoms may include a sense of being overwhelmed, feelings of anxiety, overall irritability, insecurity, nervousness, social withdrawal, loss of appetite, depression, panic attacks, exhaustion, high or low blood pressure, skin eruptions or rashes, insomnia, lack of sexual desire (sexual dysfunction), migraine, gastrointestinal difficulties (constipation or diarrhea), and for women, menstrual symptoms. It may also cause more serious conditions such as heart problems. Ayurveda is a centuries old traditional medicinal system in India. Cow is described as "Kamdhenu" (one which fulfills all the wishes) since Vedic times in Indian civilization. According to Ayurveda various cow products like urine, dung, milk, ghee and curd are used to treat various disease conditions in human beings. These five products of cow are called as Panchagavya. Panchagavya is an important component of many rituals and pooja. Many useful elements have been found in Panchagavya like Urea, Uric acid & Minerals and bioactive substances and hormones like Urokinase, Epithelium growth factor, Colony stimulating factor, Growth hormone, Erythropoietin, Gonadotropins, Kallikrin, Trypsin inhibitor, Allantoin, Anti-cancer substance, Nitrogen, Sulphur, Ammonia, Copper, Iron, Phosphate, Sodium, Potassium, Manganese, Carbolic Acid, Calcium, Salts, Vitamin A, B, C, D, E, Lactose Sugar, Enzymes, Water, Hippuric Acid, Creatinine etc. Moreover, the root, bark and leaves of *Aloe barbedansis* Mill Xanthorrhoeaceae are depurative, anthelmintic,

anti-ulcer, anti-tumours, analgesics, anti-inflammatory, hepatoprotective, immunomodulatory, wound healing & used for skin disease & leprosy etc.

Tremendous interest is generated in the therapeutic value of cow product due to the patent awarded by USFDA. This was awarded for the synergetic activity of cow urine distillate with some antibiotic and anticancer agents. But no patent awarded to other constituents of Panchagavya but there is a synergistic action of Panchagavya components either alone or combination with drug of herbal, animal or mineral origin [1], [2] .

MATERIALS AND METHODS

Collection of plant: Leaves of *Aloe barbedansis* Mill (Xanthorrhoeaceae) were collected from Narayan Baag, Jhansi (UP), India and got identified by National Vrakshayurved Research Institute (NVRI), Gwalior road, Jhansi Accession No. 21966 by Dr. Neelima Sharma (Research Officer Incharge) in May 2013. Fresh plant parts were used for macroscopical examination whereas sample which was air dried and powdered was used for microscopical studies. An exhaustive pharmacognostic study was carried out using standard methodology [3], [4].

Collection of Panchagavya: Various cow products like urine, dung, milk are collected from DRMS colony, Jhansi and curds & Ghee were prepared.

Preparation of extract: The plant leaves were dried in shade and powdered with the help of an electric grinder. Complete dried leaves powder (75 g) was packed in a soxhlet apparatus and subjected to hot continuous percolation with 40-60°C temperature for 12 hrs using 250 ml of ethanol (95% v/v) as solvent.

Drug: Alprazolam was purchased from Windlas Biotech Ltd., Dehradun

Chemicals: All Chemicals and reagents of analytical grade were purchased from Qualigen Fine Chemicals like Chloroform, Sodium hydroxide pellets, Glacial acetic acid, Ethanol, Methanol, n-Hexane, Formic acid, Silica gel G etc

Animals: Healthy albino mice of either sex weighing between 20-30 g were taken for the study. All the animals were procured from DRDE, Gwalior (M.P.). Animal were acclimatized by keeping them in animal house facility of SVCP, Jhansi. They were housed individually in polypropylene (32x24x16 cm) cages containing husk as bedding material and maintained under controlled conditions of temperature (23±2⁰C), humidity (55±5%) and 12h light and 12h dark cycle. The animals were feed with standard pellet diet and water *ad libitum*. The Committee for the Purpose of Control and Supervision of Experimental Animal (CPCSEA) of Smt. Vidyawati College of Pharmacy, Jhansi (CPCSEA Reg.No. 966/PO/a/2006/CPCSEA) was approved for conducting Anti-stress activity of different composition of Panchagavya and ethanolic extract of *Aloe barbedansis* Mill (Xanthorrhoeaceae) by using Forced Swimming Method.

Pharmacological studies

Acute toxicity studies: Healthy albino mice of either sex were orally fed with increasing doses (1, 3, 5, 7, 9) of ethanolic extract mixed with Panchagavya for 20 days. The dose upto 4g/kg (P.O.) body weight did not produce any sign of toxicity and mortality.

Experimental procedure: The animals were divided into 10 groups of 6 animals each.

Group I: PG1; Group II: PG1+ EEAB 10%; Group III: PG1+ EEAB 50%; Group IV: PG1+ EEAB 75%; Group V: PG2; Group VI: PG2+ EEAB 10%; Group VII: PG2+ EEAB 50%; Group VIII: PG2+ EEAB 75%; Group IX: Standard Alprazolam; Group X: Control Urine

Anti-stress Activity by Forced Swimming Method: All the selected animals were divided into 10 groups. In each group 6 mice were taken and they were treated with PG1, with plant extract (PG1+10% EEAB, PG1 +50% EEAB, & PG1+75% EEAB), PG2, with plant extract (PG2+10% EEAB, PG2+50% EEAB & PG2+75% EEAB) Control group with Cow Urine and Standard group with Alprazolam. All treated animal were administered at the dose level of 4mg/Kg body weight p.o. every day in the morning at 9:00 am & standard group were treated with Alprazolam at dose level 0.25mg/kg body weight. Treated animals were observed on 1st, 6th, 11th, 16th and 21st day for regular dosing.

Table No. 5 Anti-stress Activity using Forced Swimming Method (Time in Sec)

Day	PG 1	PG 1 + PE			PG 2	PG 2 + PE			Control	Standard
		10%	50%	75%		10%	50%	75%		
1	150±1.49..	214±1.13..	225±2.52..	270±23**	198±1.33..	195±1.16**	275±1.56**	280±2.21**	135±1.22	235±1.5
6	175±1.45..	255±1.37**	272±2.36**	281±1.9**	261±2.1**	245±1.15**	280±1.00**	295±1.06**	166±2.916	240±1.161
11	220±1.31..	264±1.64**	285±0.86**	290±1.25**	291±1.59**	270±0.86**	300±2.10**	306±1.65**	216±1.55	255±2.01
16	280±1.6..	282±1.82**	290±1.1**	305±1.0**	301±2.72**	295±2.69**	316±1.32**	345±2.5**	266±1.21	270±3.21
21	320±2.21**	336±1.37**	345±2.16**	358±2.43**	350±3.16**	340±.91**	374±1.64**	390±2.16**	320±1.21	310±1431

Value are mean±SEM of 6 animals. Statistical significance test for compare was done by ANOVA, followed by Dunnet's test. Comparison made between standard vs PG 1, PG 1+PE 10%, PG 1+PE50%, PG 1+PE 75%, PG 2, PG 2 +PE10%, PG 2 +PE50%, PG 2 +PE 75%. **P value <0.001, * p value<0.05.

RESULTS

On the basis of observation table (Table-5) it was found that on the 1st day of drug administration, effect of PG 1 level (p<0.01) & PG 1 + EEAB and PG 2 + EEAB (all composition) were found to be significant from control and PG1+75% EEAB and PG2+ EEAB (all composition) were found to be significant as compared to standard group. On 6th day of administration, effect of PG1 was not significant to standard at level (p<0.01) but PG 2 level (p<0.01) as well as PG 1 + EEAB and PG 2 + EEAB (all composition) were found to be significant as compared to standard group. On 11th day of administration, effect of PG 1 and PG1+ EEAB 50% & 75%) as well as PG 2 and PG2+ EEAB (50% & 75%) level (p<0.01) were found to be significant as compared to standard group and 10% composition of PG1 was found to be not significant as compared to standard group and significant as compared to control group. On 16th day after drug administration, effect of PG 1 + EEAB (50%&75%) level (p<0.01) as well as PG 2 + EEAB (all composition) were found to be significant as compared to standard and control group. On 21th day after drug administration, effect of PG 1 level (p<0.01) as well as PG 1 + EEAB and PG 2 + EEAB (all composition) was found to be significant as compared to standard and control group.

DISCUSSION

Stress is a feeling of strain & pressure which may cause many problems in the body which could be harmful. The anti-stress activity using force swimming model in the present study showed that anti-stress activity increases with time as well as percentages composition of PG and EEAB. However, it also showed that the anti stress activity of PG 1 (16th day), PG 1+ EEAB (11th day) PG 2 (on 6th, 11th 16th and 21th days), PG 2+ EEAB (10%, 50% and 75% on all days except 1th and 6th day) using force swimming model were found to be more potent than standard Alprazolam. Further, it showed that PG2 exhibited more potent anti-stress activity than PG1 which might be due to the availability of twice the number of molecule of different constituents of PG2 than that of PG1 for drug receptor interaction. The synergistic anti-stress activity of PG with EEAB from the results emended in the present study might to be due to enhanced brain Gamma Amino Butyric Acid (GABA) level of mice and decreased brain dopamine levels and plasma corticosterone levels.

CONCLUSION

The present study of synergistic action of different composition of constituents of Panchagavya (PG) as well the ethanolic extract of *Aloe barbedansis* Mill (EEAB) (Xanthorrhoeaceae) has opened new area in the field of anti-stress.

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