ORIGINAL ARTICLE ASIAN JOURNAL OF MEDICAL SCIENCES

Evaluation of toxic effects of *Coleus forskohlii* extract on various body organs of experimental animals-rats

Haritha Yadav KC[1], Yakaiah Vangoori[2], Suresh Babu Sayana[3]

[1]Assistant Research Officer, Department of Pharmacology, Captain Srinivasa Murthy Central Ayurveda Research Institute, Chennai, Tamil Nadu, [2]Associate Professor, Department of Pharmacology, Mallareddy Medical College for Women, Hyderabad, [3]Assistant Professor, Department of Pharmacology, Government Medical College, Suryapet, Telangana, India

Submission: 21-10-2022 Revision: 02-11-2022 Publication: 01-12-2022

ABSTRACT

Background: The study aimed observation of pharmacological actions and any adverse effects on various body organs of rats with *Coleus forskohlii* (CF) extract. **Aims and Objectives:** The present study was undertaken to evaluate toxic effects of CF extract in experiments animals. **Materials and Methods:** In the single-dose oral toxicity study, groups of five healthy male and female rats were dosed orally with CF extract at 2000mg/kg BW as per organization of economic cooperation and development guidelines and were observed once daily for 14 consecutive days for toxicity, general behavior, and pharmacological effects. In sub-acute oral toxicity, the test substance was administered for 28 days with daily doses of 200, 400, and 800 mg/kg body weight. In subacute oral toxicity study, groups of five male and five female rats were subjected to 10% forskolin for 28 days. Initially autonomic symptoms (gross behavioral study), followed by hematological, biochemical, and histological parameters, were evaluated. **Results:** No deaths were reported in all the toxicity studies performed. No significant changes were observed in the hematology and serum biochemistry values from the control group animals. The body weight changes and necropsy results were normal. There was no apparent progression of organ damage in any of these toxicity tests. Furthermore, CF extract did not produce any significant toxic effects in Wistar rats at 2000 mg/kg body weight and had no potential to induce organ damage. **Conclusion:** The "no observed adverse effect level" of CF extract was determined up to 2000 mg/kg.

Key words: *Coleus*; Gross behavior; Acute and sub-acute; Toxicity

Access this article online

Website: http://nepjol.info/index.php/AJMS

DOI: 10.3126/ajms.v13i12.49018

E-ISSN: 2091-0576

P-ISSN: 2467-9100

INTRODUCTION

Coleus forskohlii (CF) (*Lamiaceae*) is an aromatic herb indigenous to India. The herb received a lot of attention over the past 40 years from medical researchers as the only significant plant source of Forskolin, a bioactive *diterpene* compound with diverse pharmacological benefits. Plants of the *Coleus* species have been used as herbal medicine to treat various disorders of the cardiovascular, respiratory, gastrointestinal, and central nervous systems since ancient times.[1] The roots of this folk medicine, CF, have a long history of food use in India, in the form of pickle or condiment. In the northern parts of India, the paste of roots of the plant is used by local people as topical application on tumors and boils. In South India, the decoction of the roots is used as a tonic by the tribals of Trichigadi (Kotas). The active phytochemical Forskolin in CF was discovered in 1974 with vast array of effects on the body.

Forskolin lowers intraocular pressure (IOP) in rabbits, monkeys, and man.[2] Extensive studies were done by Majeed et al., that demonstrated successfully the IOP lowering effects of forskolin, in both animal[3] and human studies[4,5] at different strengths. Ocular toxicity of forskolin has also been well established (Unpublished results, Majeed et al.). Results

Address for Correspondence:
Dr. Yakaiah Vangoori, Associate Professor, Department of Pharmacology, Mallareddy Medical College for Women, Hyderabad, Telangana, India. **Mobile:** 8106363463. **E-mail:** mail2liraya@gmail.com

from other studies suggest that CF may help mitigate weight gain in overweight females.[6] Forskolin could possibly be used as a therapeutic agent for weight management and treatment in obese men.[7] Additionally, Forskolin is more effective than sodium cromoglycate in preventing asthma attacks in patients with mild persistent or moderate persistent asthma.[8]

Despite its huge beneficial effects, its toxicity data have not been reported in any scientific literature till date. The present study was designed to investigate the acute, subacute, and chronic toxicity of CF extract standardized for 10% forskolin in male and female Wistar rats.

Aims and objectives

Study was aimed to evaluate the toxic profile of coleus forskohlii in rats.

MATERIALS AND METHODS

Preparation of CF root extract (CFE)

The preparation procedure to obtain CFE is covered in the following sections:

Collection, identification, and authentication of CF

The fresh tuberous root of CF was procured from contract farmer at Tiruvannamalai District of Tamil Nadu State. The plant CF was authenticated by Dr. P. Ramachandra Reddy, Professor of Medicinal Botany, Department of Botany, Osmania University, Hyderabad, Telengana. IAEC permission was take before starting study, IAEC approval NO; pharma/OMC/20/2011.

Washing, drying, and milling of the plant material

The tuberous roots were washed with water to remove adhering dirt, cut into pieces, and dried under shade. The dried roots of CF roots were made into coarse powder using a pulveriser for decreasing the moisture content. The coarse powder was stored in well closed-light resistant container until further use.

Preparation of root extract

Coleus root material was subjected to extraction using Soxhlet apparatus. At 600°C, the pulverized material was subjected to the Soxhlet apparatus by the continuous percolation extraction process. Using standard protocols given by Trease and Evans,[9,10] the roots of CF was treated with *Toluene*, which was used as a solvent for extraction in 1:4 (raw materials to solvent) ratio, till spent assay was nil. Extracted solvent was concentrated through distillation. After concentrating the toluene extract, hexane was added to the concentrate for purification. Concentrated extract was mixed well with hexane and kept for settling for 24 h.

Toxicity studies – design of the experiments

Acute oral toxicity study

Acute oral toxicity study for the CFE dissolved in 0.5% Carboxy methyl cellulose (CMC) was conducted in both male and female Wistar albino rats as per the World Health Organization guideline-9 and the organization of economic cooperation and development (OECD) guideline for testing of chemicals.[11,12]

Animal preparation

Healthy young Wistar Albino rats weighing 150–200 g adult were acclimatized to the laboratory conditions for 7 days before test before assigning the animals to treatment groups. They had free access to food and water and were maintained under standard laboratory conditions which included 12-h light-dark cycle and temperature of 28–30°C.

Animal groups and number of animals

Male and female rats were caged separately in polypropylene cages. Four groups of rats consisting of five males and five females in each group were used for the study.

Administration of doses

The animals were fasted overnight before the administration of the substance. The 2000 mg/10 ml CFE in 0.5% CMC was administered to rats at single oral dose of 2000 mg/kg body weight by oral gavage, and 0.5% CMC was also given to the control group of rats.

Observation of animals for toxicity symptoms

Careful general clinical observations were made every day. All the animals were observed for effects on skin and face, eyes, mucous membranes, respiratory and circulatory systems, autonomic change such as salivation, central nervous system effects including tremors and convulsions, and changes in the level of activity, gait, posture, reactivity to handling or sensory stimuli, and altered strength, health conditions, and mortality. Animals were observed individually after dosing at least once during the first 30 min, periodically during the first 24 h, with special attention given during the first 4 h and daily thereafter. The survival rats were weighed daily and observed further for clinical signs of toxicity for up to 14 days. After the experimental period, rats of both groups were sacrificed and their internal organs such as heart, lungs, livers, kidneys, spleen, adrenals, sex organs, and brain were examined for altered gross structural changes such as hemorrhage (gross necropsy).[13]

Repeated dose 28 days oral toxicity study

Toxicity study was conducted with CF root extract according to OECD guidelines. Acute and subacute study was done in 28 days in male and female rats.[12]

Selection of doses

The dose was selected on the basis of the previous studies and acute toxicity study where the highest dose was chosen

with the aim of inducing toxicity but not death or reverse suffering. Thereafter, a descending sequence of dose was selected with a view to demonstrate any dose related response and no observable adverse effects level (NOAEL) at the lowest dose. Two to four-fold descending dose intervals were selected as per the guidelines.

Observations

The observation period was for 28 days. Animals in a satellite group were scheduled for follow-up observations and were kept for at least 14 days without treatment to detect delayed occurrence, or persistence of, or recovery from toxic effects. General clinical observations were made at least once a day, preferably at the same time each day and considering the peak period of anticipated effects after dosing. The health condition of the animals was recorded. At least twice daily, all animals are observed for morbidity and mortality. Once before the first exposure (to allow for within-subject comparisons), and at least once a week thereafter, detailed clinical observations were made in all animals. These observations were made outside the home cage in a standard arena and preferably at the same time of day on each occasion. Signs noted include, but not be limited to, changes in skin, fur, eyes, mucous membranes, occurrence of secretions and excretions, and autonomic activity (e.g., lacrimation, polierection, pupil size, and unusual respiratory pattern). Changes in gait, posture, and response to handling as well as the presence of clonic or tonic movements, stereotypes (e.g., excessive grooming and repetitive circling) or bizarre behavior (e.g., self-mutilation and walking backward).[13]

RESULTS AND DISCUSSION

Acute toxicity studies (Gross behavioral study)

After the administration of 2000 mg/kg BW of CF extract to Wister rats, they were initially observed for their reaction or behavior toward the drug for every 30 min. The rats showed some behavioral changes, 120 min after oral administration. The observation for clinical signs continued further for every hour starting from the 1st h to 24 h of drug administration. These changes included slow response to external stimuli, reduction of mobility and aggression, slight excitability sketching, and sluggishness. However, after 24 h, all the changes observed before disappeared. This process of observation was carried out daily for 14 days. No toxic signs and death were recorded in any animal of both the sexes.

All the animals in treated group were alive up to 14 days after administration of the plant extract. The behavioral changes were observed from 1st day to 14th day in control and treated groups. Both the groups were normal, and no changes were observed in behavior, sleep, eyes, salivation, diarrhea like problems, skin and hair loss, lethargy, and food consumption and water intake. Moreover, body weights were also very similar in normal control and test group. Hence, the CFE can be classified in the category of substances with low toxicity.

Repeated dose sub-acute toxicity study is fundamental requirement for the assessment of toxicological profile of any drug. Repeated administration of drugs gives information on the tolerability and nature of toxic effects due to cumulative action of drugs. Toxicological investigations aim to determine dose related toxic effects of drugs on specific organs such as bone marrow, heart, kidneys, and liver.

Because many of the drugs are associated with toxic effects of specific vital organs and hence it was decided to investigate sub-acute toxicity study for understanding the details of the toxicological properties. Sub-acute toxicity study involves repeated daily dosing of the animals and various investigations related with body weight, feed intake, biochemical, and hematological parameters. Administration of CF extracts did not show any mortality during the study period. Various parameters investigated after 28 days of treatment were change in weight of vital organs in different groups of animals, hematological parameters, biochemical parameters, and histological analysis of vital organs.

Effect on biochemical parameters

Apart from the morphological and histological analysis, toxicity study of liver and kidney generally involves biochemical testing of functional markers of liver and kidneys.[14] Hence levels of various markers in serum were determined for understanding toxic effect of CF extract.

Serum alanine transaminase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) are the marker enzymes are found to be elevated in hepatotoxicity along with elevations in total cholesterol and triglycerides as a result of loss of structural and integrity of hepatocytes.[15,16]

The levels of markers enzyme ALT were not significantly different among the animals treated with different doses of CF extract compared to the control animals. The levels of AST were also found to be statistically indifferent in the animals treated with different doses of CF extract as compared to the control animals. Serum ALP levels of the animals treated with different doses of CF extract were not significantly different from that of control animals Tables 3 and 4.

Bilirubin is a bile pigment that is formed in the process of breakdown of hemoglobin; hyperbilirubunaemia is always

associated with the hepatic damage, many hepatotoxic and irritant substances cause elevations in serum bilirubin levels.[17] However, chronic administration of CF extracts does not caused changes in bilirubin levels as compared to the normal rats which indicate absence of a toxic effect on liver.

Table 1: Symptoms recorded during acute toxicity studies of the Coleus forskohlii root extract of the albino male rats

Observation	Control group	Treated group
Behavior	Normal	Normal
Skin and fur	Normal	Normal
Sleep	Normal	Normal
Eyes	Normal	Normal
Salivation	Normal	Normal
Diarrhea	Normal	Normal
Lethargy	Normal	Normal

Table 2: Symptoms recorded during acute toxicity studies of the Coleus forskohlii root extract of the albino female rats

Observation	Control group	Treated group
Behavior	Normal	Normal
Skin and fur	Normal	Normal
Sleep	Normal	Normal
Eyes	Normal	Normal
Salivation	Normal	Normal
Diarrhea	Normal	Normal
Lethargy	Normal	Normal

Similar to the bilirubin, the level of serum cholesterol and triglycerides is frequently determined as elevated values of both cholesterol and triglycerides are found in hepatotoxicity.[18] Result in Tables 3 and 4 indicates that the level of cholesterol in animals of Group III (CFE 200 mg/kg b.w. p.o.), Group IV (CFE 400 mg/kg b.w.p.o.) and Group V was not significantly different from Group I (control) animals. Similarly, the levels of triglycerides were also found to be statistically indifferent in animals of Group III (CFE 200 mg/kg), Group IV (CFE 400 mg/kg b.w. p.o.), and Group V group; as compared to Group I (control) animals. Toxic effects on kidneys are associated with elevations of serum urea, uric acid, and creatinine;[19] however, the serum levels of urea, uric acid, and creatinine in the present study do not differ significantly from control animals indicating that the chronic administration of CF do not cause any renal toxicity.

Effect on organ weights

At autopsy, the organ such as thymus, heart, liver, spleen, kidneys, brain, testes, adrenals, and epididymis were collected by separating them from adhering tissues and weighed individually. The organ weights for treated and control group of male and female rats are presented in Tables 5-7.

Organ weights in male rats

There were no significant (P>0.05) changes in weight of thymus, heart, liver, spleen, kidneys, brain, testes, adrenals, and epididymis in intermediate dose and high

Table 3: Effect of *Coleus forskohlii* root extract of low dose (200 mg/kg bw), intermediate dose (400 mg/KG BW), and high dose (800 mg/KG BW) on biochemical parameters after 28 days repeated dose administration in male rats

Parameters	Control		Low dose		Intermediate dose		High dose	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
AST	68	15.19	66.2	13.85	62.09	22.09	69.6	15.36
ALP	43.6	11.97	49.4	4.39	45.67	5.67	46	4.06
Bilirubin	0.63	0.16	0.69	0.04	0.61	0.01	0.62	0.04
Glucose	83.6	4.83	88.2	4.7	88.75	5.75	84.2	5.22
Total Cholesterol	67.4	3.11	67.8	5.63	68.23	8.23	61.2	3.94
Triglycerides	109.4	26.16	106	24.51	109.91	29.91	109.4	23.73

ALT: Alanine transaminase, AST: Aspartate aminotransferase

Table 4: Effect of *Coleus forskohlii* root extract of low dose (200 mg/kg BW), intermediate Dose (400 mg/KG BW) and high dose (800 mg/KG BW) on biochemical parameters after 28 days repeated dose administration in female rats

Parameters/Groups	Control		Low dose		Intermediate dose		High dose	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
AST	62.2	10.55	60.4	19.01	68	16.9	60.6	13.94
ALP	40.6	10.88	47.2	12.44	49.6	5.03	41.2	9.07
Bilirubin	0.64	0.08	0.68	0.1	0.58	0.03	6.2	0.04
Glucose	96	2.08	85.8	8.58	83.4	2.95	88	2.35
Total Cholesterol	67.8	8.87	50.6	9.26	80.8	6.51	67.6	5.13
Triglycerides	107.2	29.96	101	22.75	108.6	26.41	108	25.64

Values mentioned in results are average of 5 readings in each group and standard deviation for a group, statistically significant differences are indicated by asterisks (*P<0.05, compared with control group), CFE: *Coleus forskohlii* root extract, ALT: Alanine transaminase, AST: Aspartate aminotransferase

Table 5: Effect of Coleus forskohlii root extract of low dose (200 mg/kg bw), intermediate dose (400 mg/kg BW), and high dose (800 mg/kg BW) on organ weights after 28 days repeated dose administration in male rats

Group	Dose (mg/kg)	Adrenal gland	Kidney	Testes	Epididymis
Ia	Control	0.022±0.002	1.0212±0.00	1.2032±0.16	0.357±0.03
IIa	Control (REV)	0.027±0.003	1.058±0.60	1.1996±0.10	0.3062±0.11
IIIa	CFE (200)	0.0218±0.001	1.1414±0.07	1.3208±0.38	0.3984±0.06
IVa	CFE (400)	0.0244±0.003	0.9484±0.17	1.2106±0.65	0.3404±0.04
Va	CFE (800)	0.0208±0.001	1.0598±0.06	1.0474±0.06	0.4118±0.06
VIa	800 (REV)	0.0236±0.004	1.0022±0.05	1.1182±0.11	0.3738±0.01

Values mentioned in results are average of five readings in each group and standard deviation for a group, statistically significant differences are indicated by asterisks (*P<0.05, compared with control group), CFE: *Coleus forskohlii* root extract

Table 6: Effect of Coleus forskohlii root extract of low dose (200 mg/kg BW), intermediate dose (400 mg/kg BW), and high dose (800 mg/kg BW) on organ weights after 28 days repeated dose administration in male rats

Group	Dose (mg/kg)	Thymus	Heart	Spleen	Brain	Liver
Ia	Control	0.474±0.02	0.474±0.02	0.8206±0.13	1.9446±0.04	9.6362±0.84
IIa	Control (REV)	0.445±0.04	0.445±0.04	0.993±0.12	1.8664±0.11	9.7312±0.96
IIIa	CFE (200)	0.4976±0.02	0.4976±0.02	0.906±0.04	1.9474±0.11	9.6198±0.45
IVa	CFE (400)	0.4324±0.07	0.4324±0.07	0.9592±0.08	1.843±0.08	8.6866±0.96
Va	CFE (800)	0.4384±0.07	0.4384±0.07	0.9634±0.06	1.8026±0.10	8.7776±0.25
VIa	800 (REV)	0.4528±0.04	0.4528±0.04	1.0656±0.10	1.8664±0.11	10.555±0.45

Values mentioned in results are average of five readings in each group and standard deviation for a group, statistically significant differences are indicated by asterisks. (*P<0.05, compared with control group), CFE: *Coleus forskohlii* root extract

Table 7: Effect of *Coleus forskohlii* root extract of low dose (200 mg/kg BW), intermediate dose (400 mg/kg BW), and high dose (800 mg/kg BW) on organ weights after 28 days repeated dose administration in female rats

Group	Dose (mg/kg BW)	Adrenal	Kidney	Ovary
Ib	Control	0.0206±0.003	0.7964±0.04	0.0504±0.009
IIb	Control (REV)	0.0234±0.002	0.7826±0.031	0.0472±0.001
IIIb	CFE (200)	0.0206±0.002	0.746±0.136	0.0452±0.004
IVb	CFE (400)	0.023±0.002	0.811±0.113	0.0526±0.022
Vb	CFE (800)	0.0206±0.0008	0.59±0.215	0.047±0.007
VIb	800 (REV)	0.0226±0.004	0.45±0.030	0.0402±0.002

Values mentioned in results are average of five readings in each group and standard deviation for a group, statistically significant differences are indicated by asterisks. (*P<0.05, compared with control group), CFE: *Coleus forskohlii* root extract

Table 8: Effect of Coleus forskohlii root extract of low dose (200 mg/kg BW), intermediate dose (400 mg/kg BW), and high dose (800 mg/kg BW) on organ weights after 28 days repeated dose administration in female rats

Group	Dose (mg/kg)	Thymus	Heart	Spleen	Brain	Liver
Ib	Control	0.463±0.048	0.463±0.048	0.8206±0.13	1.8306±0.05	6.428±0.28
IIb	Control (R)	0.4444±0.049	0.4444±0.049	0.6698±0.04	1.8268±0.10	8.0444±1.16
IIIb	CFE (200)	0.4734±0.052	0.4734±0.052	0.6698±0.03	1.8268±0.10	8.0428±1.16
IVb	CFE (400)	0.4784±0.037	0.4784±0.037	0.6438±0.08	1.7826±0.171	7.7236±0.99
Vb	CFE (800)	0.427±0.100	0.427±0.100	0.8444±0.09	1.747±0.09	6.3812±0.27
VIb	800 (REV)	0.4716±0.033	0.4716±0.033	0.8464±0.09	1.8684±0.05	6.8128±0.22

Values mentioned in results are average of five readings in each group and standard deviation for a group, statistically significant differences are indicated by asterisks (*P<0.05, compared with control group), CFE: *Coleus forskohlii* root extract

dose groups of male rats compared to Control group rats (Tables 5 and 6). Satellite high dose group administered with 800 mg/kg body weight of CFE showed normal recovery and did not show any significant (P>0.05) changes in organ weights compared to satellite control group values of male rats with 14 days of post withdrawal period.

Organ weights in female rats

There were no significant (P>0.05) changes in weight of thymus, heart, liver, spleen, kidneys, brain, testes, adrenals, and epididymis in intermediate dose and high dose groups of female rats compared to control group rats (Tables 5-8). Satellite high dose group administered with 800 mg/kg

body weight of CFE showed normal recovery and did not show any significant (P>0.05) changes in organ weights compared to satellite control group values of male rats with 14 days of post withdrawal period.

Histological study

Microscopic observation of histological slides liver did not reveal any toxicity related changes such as fatty infiltration, changes in cell size, or degradation of hepatocytes.

There were no toxic changes in the tissue of heart with the chronic treatment with different doses of extract of CF as compared to the control animals, similarly the microscopic observation of histology of kidney did not reveal any toxicity-related changes and appeared nearly normal in animals treated with different doses of extract of CF as compared to the control animals.

Taken together, results of this study indicate that the acute and subacute oral administrations of different doses of extract of CF produced no mortalities. There were no significant toxic effects on body weight of animals and there were no signs of toxicities of the vital organs such as heart, liver, and kidneys as evident from their gross appearance, weight, and histological analysis. The extracts do not have any toxic effects on functional capacity of kidneys and liver as seen from the biochemical analysis. There was no harmful effect observed on bone marrow and hematocrit in rats and can be generally regarded as safe for medicinal use.

Limitations of the study

Chronic study should have been done with other animal models. Molecular docking study can be done.

CONCLUSION

After collection of plant material, extract was prepared from roots of coleus forskohlii by using soxhlet apparatus. The extract was subjected to do acute and subacute, 2000mg/kg BW study in pre-designed animal models. In acute study, animals were fed with CF extract for 14 days in a dose of 2000mg/kg. No observational symptoms were appeared in all groups of rats. When it was considered as safe in acute study, chronic study was conducted for 28 days as per OECD guidelines. The extract showed no signs of toxicity in all the body organs and there are no alterations in physiological actions of any system. Hence, it could be concluded that a repeated oral exposure to this extract up to 2000 mg/kg b.wt/day does not produce any toxic effects and may be treated as NOAEL under the test conditions employed.

ACKNOWLEDGMENT

The author thanking UGC for providing PhD fellowship during the study and SVS Medical college, Mahabubnagar, for providing good laboratory facilities for conducting the study.

REFERENCES

1. Agarwal KC, Parks RE Jr. Forskolin: A potential antimetastatic agent. Int J Cancer. 1983;32(6):801-804. https://doi.org/10.1002/ijc.2910320622
2. Caprioli, J and Sears M. Forskolin lowers intraocular pressure in rabbits, monkeys, and man. Lancet. 1983;1(8331):958-960. https://doi.org/10.1016/s0140-6736(83)92084-6
3. Majeed M, Nagabhushanam K, Natarajan S, Vaidyanathan P and Karri SK. Intraocular pressure lowering activity of ocufors® (forskolin 0.15% w/v ophthalmic solution) in water loaded New Zealand white rabbits. Int J Pharm Bio Sci. 2014;5(4):328-336.
4. Majeed M, Nagabhushanam K, Natarajan S, Vaidyanathan P and Karri SK. A double-blind, randomized clinical trial to evaluate the efficacy and safety of forskolin eye drops 1% in the treatment of open angle glaucoma. J Clin Trials. 2014;4(5):1-6. https://doi.org/10.4172/2167-0870.1000184
5. Majeed M, Nagabhushanam K, Natarajan S, Vaidyanathan P, Karri SK and Jose JA. Efficacy and safety of 1% forskolin eye drops in open angle glaucoma-an open label study. Saudi J Ophthalmol. 2015;29(3):197-200. https://doi.org/10.1016/j.sjopt.2015.02.003
6. Henderson S, Magu B, Rasmussen C, Lancaster S, Kerksick C, Smith P, et al. Effects of *Coleus forskohlii* supplementation on body composition and hematological profiles in mildly overweight women. J Int Soc Sports Nutr. 2005;2(2):54-62. https://doi.org/10.1186/1550-2783-2-2-54
7. Godard MP, Johnson BA and Richmond SR. Body composition and hormonal adaptations associated with forskolin consumption in overweight and obese men. Obes Res. 2005;13(8): 1335-1343. https://doi.org/10.1038/oby.2005.162
8. González-Sánchez R, Trujillo X, Trujillo-Hernández B, Vásquez C, Huerta M and Elizalde A. Forskolin versus sodium cromoglycate for prevention of asthma attacks: A single-blinded clinical trial. J Int Med Res. 2006;34(2):200-207. https://doi.org/10.1177/147323000603400210
9. Trease G and Evans W. Textbook of Pharmacognosy. London: Balliere-Tindal; 1979.
10. Semiz A and Sen A. Antioxidant and chemoprotective properties of *Momordica Charantia* L (bitter melon) fruit extract. Afr J Biotechnol. 2007;6(3):273-277.
11. Health Effects Test Guidelines, Acute oral toxicity (Computer Program). OPPTS 870, 1100 United States Office of Prevention, Pesticides and Toxic Substances Environmental Protection Agency (7101). Available from: https://www.epa.gov/opptsfrs/home/guideline.htm [Last accessed on 2004 Jun 05].
12. OECD. OECD/OCDE: OECD Guidelines for the Testing of Chemicals. Paris: Organisation for Economic Co-Operation and Development; 2008.
13. Parasuraman S. Toxicological screening. J Pharmacol Pharmacother. 2011;2(2):74-79. https://doi.org/10.4103/0976-500X.81895
14. Baskaran C, Rathabai V, Sivamani P and Thiagarajan V. Antimicrobial activity of various root extracts of *Coleus forskohlii*.

Int J Curr Sci. 2011;1:78-84.

15. Shan Y, Wang X, Zhou X, Kong L and Niwa M. Chemical and pharmaceutical bulletin. Chem Pharm Bull (Tokyo). 2007;55(12):376-381.
16. Khatun S, Cakilcioglu U, Chatterjee NC. Phytochemical constituents vis-a-vis histochemical localization of forskolin in a medicinal plant *Coleus forsksohlii* Briq. J Med Plants Res. 2011;5(5):711-718.
17. Murugesan S, Rajeshkannan C, Sumathi R, Manivachakam P and Babu DS. Bioactivity of root hexane extract of *Coleus forskohlii* Briq. Labiatae : GC/MS/MS characterization and identification. Eur J Exp Biol. 2012;2(5):1469-1473.
18. Patil S, Hulamani NC and Rokhade AK. Performance of genotypes of *Coleus forskohlii* Briq. for growth, yield and essential oil content. Indian Perfum. 2001;45:17-21.
19. Atulkar P, Thakur R and Singh P. Preliminary phytochemical analysis of root extracts of *Coleus forskohlii* Briq. Int J Adv Res. 2015;3(12):1145-1150.

Authors' Contributions:
HYKC- Concept, and design of the study, results interpretation, review of the literature, and preparing the first draft of the manuscript; **YV-** Concept, and design of the study, results interpretation, review of the literature, and preparing the first draft of the manuscript; and **SBS-** Concept and design of the study, statistical analysis and interpretation, and revision of the manuscript.

Work attributed to:
Captain Srinivasa Murthy Central Ayurveda Research Institute, Chennai. Mallareddy Medical College for Women (MRMCW), Suram X-road, Jidimetla. Hyderabad. Telangana and Government Medical College, Suryapet, Telangana, India.

ORCID ID:
Dr. Haritha Yadav KC- https://orcid.org/0000-0003-0893-2243
Dr. Suresh Babu Sayana - https://orcid.org/0000-0003-4971-4007

Source of Funding: None, **Conflicts of Interest:** None.