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Research Article

LAXATIVE AND DIURETIC PROPERTY OF ETHANOLIC EXTRACT OF LEAVES OF *ALOCASIA MACRORRHIZA* LINN. ON EXPERMENTAL ALBINO RATS

Uddin Sheikh Mubeen*, Misra Vimlesh, Banerjee Santanu[1]
Institute of Pharmaceutical Research, GLA University, Mathura, Uttar Pradesh, India
[1]S.R.L.T.Institute of Pharmaceutical science and technology, Etawah, Uttar Pradesh, India

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*E-mail: mubeen.pathan03@gmail.com

ABSTRACT
The present study was carried out to evaluate the laxative and diuretic effect of *Alocasia macrorrhiza* leaves extract in rats. The ethanolic extract was found to produce significantly laxative activity in dose dependent manner. On other hand diuretic and natriuretic activities were carried out by administration of normal saline along with the treatment modules. The volume of urine (in ml) and the Na+ and K+ content in the urine were measured. The ethanolic extract of 100, 200 and 400 mg / kg, produced significant laxative, diuretic and natriuretic activity. Presence of different phytoconstituent in ethanolic extract of *Alocasia macrorrhiza* may be responsible for the specific activities. Overall, the extract was found to be significant laxative and diuretic activity.
KEY WORDS: *Alocasia macrorrhiza*, acute toxicity study, Diuretic activity, Laxative activity.

INTRODUCTION

Herbal medicines derived from plant extracts are being increasingly utilized to treat a variety of disease. There have little knowledge about their pharmacological action[1]. Medicinal plants are important source of unknown chemical substance with potential therapeutic effect[2]. Constipation also known as costiveness refers to bowel movement that is infrequent and/or hard to pass[3]. Constipation causes are two Types: obstructed defecation and colonic slow transit (hypo mobility)[4]. Constipation is a highly prevalent, after chronic gastrointestinal disorder that affects adult[5, 6]. Laxative are among the most widely prescribed drug for the treatment of constipation[7]. Diuretics are the drug that increases urine volume; clinically useful diuretics also increase the rate of excretion of Na^+ (natriuresis), Cl^- and water[8]. Diuretics are the first line drug for the treatment of hypertension. Diuretic has also major impact on the understanding of renal physiology[9]. *Alocasia macrorrhiza* (L)schott (family-Araceae, syn. *Alocasia indica*(L.) schott, *Alocasia macrorrhizos*) commonly known as manakachu and sholakachu in Bangladesh[10] . *Alocasia macrorrhiza* are naturally grown in marshy land of tropical area in India, Bangladesh, China and south Africa[11]. The ethanolic extract of the leaves of the plant are used as antioxidant, anti-inflammatory, antinociceptive[12], antimicrobial[13], antidiarrheal[14], free radical scavenging activity[15] and antiprotozoal. The hydroalcoholic extract of the leaves are used as hepatoprotective[16] and anthelmentic activity[17]. The juice of the leaves given in colic, constipation, digestive, laxative, diuretic, astringent and rheumatic arthritis patient traditionally[18]. *Alocasia macrorrhiza* has antifungal[19] and antitumour properties[20]. Petioles contain HCN (up to 0.018%) used for toothache and their juice used for coughs[21]. Rhizomes are used for abdominal pain, vomiting and reduce elevated blood glucose level traditionally[22].

The plant contain The plant contains Oxalic acid, flavonoids, cholesterol, amino acids, gallic acid, mallic acid, ascorbic acid, succinic acid, glucose cyanogenetic glycosides, alocasin, fructose, sucrose, betalectins[13], triglochinin, ceramide[23]isotriglochinin,β-glucosidases,phytosterol like compound[24], ergosterol, campesterol, stigmasterol, β-sitosterol and clionasterol[25]. The spadix contain following amino acid: leucine, α-alanine, proline, glutamic acid, glycine, valine, aspartic acid, γ-amino butyric acid, threonine and serine. Small amount of lysine, cystine, arginine, histidine, phenylalanine, tyrosine, glutamine and asparagines are also present. The present study aims at exploring the detail of laxative and diuretic action of ethanolic extract of *Alocasia macrorrhiza* leaves.

MATERIAL AND METHOD

Collection of plants

Fresh leaves of *Alocasia macrorrhiza* were collected from different places of Mahoba(Uttar Pradesh). Leaves of *Alocasia macrorrhiza* were authenticated and specimen was deposited at birbal sahni institute of paleobotany, lucknow

Preparation of extract

The leaves were dried in shade at room temperature and crushed coarse powder. The powder was passed through sieve number 14 to obtain a uniform sized powder; the powder was loaded in soxhlet apparatus and extraction process complete 30 cycles with ethyl alcohol (95%). After extraction the solvent distilled off by using vacuumed distillation. Extract was concentrated on water bath to dry residue[26].

Experiment animal

Wistar albino rats of either sex, weighing 170-200g provided by CDRI lab of lucknow, India. Before initiation of experiment, rats were acclimatized for a period of 7 day. Standard environment condition was maintained. The animal was allowed to standard pellet diet and tap water ad libitum. The experiment protocol has been approved by the institutional animal ethics committee and by regulatory body of the government.

Acute toxicity study

The acute toxicity of ethanolic leaf extracts of plant *Alocasia macrorrhiza* was determined using Swiss albino mice as per OECD 425 guideline, the animals were observed continuously for the behavioral changes for the first 2, 4 h and then observed for mortality if any, after 24 h.

Evaluation of laxative activity

Laxative activity was performed according to capasso et.al. On wistar albino rats of either sex, fasted for 12 hours before the experiment, but with water provided ad libitum. Rats were divided in five groups, each group consisting six rats.

The first group of animal, received normal saline (25ml/kg, p.o.), second group of animals received Agar-agar (300mg/kg, p.o.) the third, fourth and fifth groups of animals received simultaneously (100,200,400 mg/kg, p.o.) ethanolic extract of *Alocasia macrorrhiza*. After administration the animal were placed in a plastic container suitable for collection of faces. After 8 hours of drug administration, the faces were collected and weight. Thereafter, food and water were given to all rats and faecal outputs were again weight after a period of 16 hours[4].

Evaluation of diuretic activity

Lipschitz test described by Lipschitz *et.al.* (1943) was employed for assessment of diurectic activity. In this method, wistar albino rats of either sex weight 150 to 200 gm were used. The rats were divided five groups of six animals each. The animals were fasted for 24 hours prior to the experiment and water was given ad libitum during fasting. The first group of animal, received normal saline (25ml/kg, p.o.), second group of animals received furosemide (20mg/kg, p.o.), third, fourth and fifth groups of animals received simultaneously (100,200,400 mg/kg, p.o.) ethanolic extract of *Alocasia macrorrhiza.* After administration the animal were placed in a metabolic cage (2 per cage), specially designed to separate urine and faces, and kept at 20ºc±0.5ºc. The volume of urine collected was measured at the end of 5 hours. During this period, no food and water was made available to animals. The parameter was taken volume of urine, electrolytes (Na^+, k^+, Cl^-) were estimated in urine for assessment of diuretic activity. The Na^+, k^+, estimated was carried out using flame photometry. The Cl^- ion concentration was estimated by titration with 0.02N $AgNO_3$ using 5%potassium chromate solution as indicator [27].

Statistical analysis

All results are expressed as mean ±standard error. The data was analyzed using one ways of analysis of variance (ANOVA). The statistical significance of the difference of the means was evaluated by Dunnet's test.

RESULT

The preliminary photochemical test revealed the presence of flavonoid, cholesterol, amino acids, glycoside and alkaloid in the ethanolic extract of *Alocasia macrorrhiza*. In laxative study, the different doses of the extract showed dose dependant increase in fecal output of rats when compared to the control group. However the test extract at lower dose (100mg/kg) failed to show the effect of laxative. The effect of *Alocasia macrorrhiza* at dose of 200 and 400 mg/kg, p.o. increased significantly fecal output of rats compared to control group. The laxative activity demonstrated by the test extract of 400mg/kg was significantly lesser than standard drug Agar-agar (300mg/kg,p.o.). The result is complied in the table 2. The present study revealed that, ethanolic extract of *Alocasia macrorrhiza* significantly increases the urinary output as well as urinary ion concentration at higher doses. However the test extract at lower dose (100mg/kg P.O.,) failed to do so. The ethanolic extract was found to produce significant increase in excretion of Na^+, k^+, Cl^- ions at the higher dose tested (400mg/kg p.o.). The order of activity of increase of urinary output was 400mg>200mg>100mg .The diuretic activity demonstrated by the test extract of 400mg/kg was significantly lesser than standard drug furosemide (20mg/kg). The result is complied in the table 1.

DISCUSSION

In the present study, we demonstrated the laxative effect of *Alocasia macrorrhiza* in a rat model of low-fiber diet-induced constipation. Maintaining a low fiber diet for 5 weeks significantly decreased stool frequency, weight and water content. A single administration of *Alocasia macrorrhiza* at 400mg/kg also significantly accelerated stool frequency, weight, and water content. Multiple administrations of *Alocasia macrorrhiza* at 200 and 400 mg/kg also significantly increased the frequency and weight of stools, and at 100-400 mg/kg, multiple administrations significantly increased stool water content. A single treatment of *Alocasia macrorrhiza* at 100 and 200 mg/kg did not show efficacy, but repeated treatment of *Alocasia macrorrhiza* at 200 mg/kg showed a significant increase in stool weight and stool frequency. These results indicate that *Alocasia macrorrhiza* ameliorated low-fiber diet-induced constipation in rats; therefore, *Alocasia macrorrhiza* may be suitable for human patients suffering from constipation due to their diet style. Diuretics are one of the groups of drugs used for the treatment of hypertension. Diuretics relieve pulmonary congestion and peripheral edema. They decrease plasma volume and subsequently venous return to heart. This decreases cardiac work load, oxygen demand and plasma volume, thus lowering blood pressure. They are the first line of drugs in the treatment of mild to moderate hypertension along with sodium restriction in the diet. The current study evaluated the diuretic potential of *Alocasia macrorrhiza* leaves in Wistar albino rats. The purpose of the present study was to establish the scientific basis for the traditional and the reported folk use of *Alocasia macrorrhiza* for diuresis. Ethanolic extract of *Alocasia macrorrhiza* leaves significantly increased the urinary output as well as urinary electrolyte concentration at a dose of 400mg/kg, p.o. but the effect was found to be less potent in increasing the urinary output when compared with the reference drug. Further the ethanolic extract of *Alocasia macrorrhiza* leaves were found to be more effective in enhancing urinary electrolyte concentration for all three ions tested (Na^+, K^+, Cl^-).

The increase in the ratio of concentration of excreted Na^+ and K^+ ion indicated that the extract increasing Na^+ ion excretion to a greater extent than K^+ ion, which is very essential requirement of an ideal diuretic with lesser hyperkalaemic side effect.

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Table 1 : Diuretic Effect of ethanolic extracts of *Alocasia macrorrhiza* in rats.

Treatment	Urine volume (ml/100g/5h)	Na^+(mmol/L)	K^+(mmol/L)	Cl^-(mmol/L)	Na^+/K^+
control	4.62±0.60	104.80±1.30	92.20±0.82	58.32±5.28	1.137
standard	8.26±0.46***	125.92±0.16**	62.01±2.11***	78.25±1.70*	2.031
100mg/kg ethanolic extract(n=6)	5.73±0.38^{ns}	112.58±3.50^{ns}	89.30±2.90^{ns}	55.30±3.25	1.261
200mg/kg ethanolic extract(n=6)	6.90±0.32*	118.20±3.12**	78.82±1.30***	60.83±5.37	1.499
400mg/kg ethanolic extract.(n=6)	7.68±0.60***	123.80±2.65***	72.40±1.30***	62.48±5.65	1.709

Values are given as mean±S.E.M;n=6, *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$ considered for significance,(ANOVA followed by Dunnett's test) .

Table 2: Laxative activity of ethanolic extract of *Alocasia macrorrhiza* in rats

Treatment	Fecal output	
	8 hours	8-16hours
control	0.751±0.42	1.609±0.65
Agar-agar	5.086±1.12**	5.418±0.60**
100mg/kg ethanolic extract(n=6)	0.890±0.16	1.192±0.29
200mg/kg ethanolic extract(n=6)	3.705±0.78*	4.734±0.11
400mg/kg ethanolic extract(n=6)	4.825±0.91**	5.217±0.58**

Value are expressed as mean±S.E.M ;n=6, *$P < 0.05$ compared to control group;and; **$P < 0.01$ compared to control group.

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