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Preclinical Evaluation Of Antiurolithiatic Activity Of *Pistia stratiotes* On Sodium Oxalate Induced Urolithiasis.

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ABSTRACT

The present study was undertaken to evaluate the antiurolithiatic activity of the *Pistia stratiotes* (PS). The antiurolithiatic action of methanolic extract of *Pistia stratiotes* (MEPS) was studied. MEPS was given every day orally at doses of 200 and 400 mg/kg for 10 days to albino rats to evaluate the activity against sodium oxalate mediated urolithiasis, with the reference standard of Cystone (500 mg/kg, p.o.). The effect of the extract on various parameters and histopathological examinations were studied. Showed that the plant *Pistia stratiotes* was showed the significant antiurolithiatic activity against sodium oxalate crystals, which could be a potential source for the treatment of renal stone disease.

Keywords: Urolithiasis, *Pistia stratiotes*, Sodium oxalate and Cystone

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INTRODUCTION

The usage of herbal medicine is one of the oldest forms of health care system. It has been used in all cultures throughout the history[1]. About 75-80% of world population is depending herbal medicine for primary health care[2]. Diseases are born along with man's birth, drug came into existence very long back to prevent and cure the disease. So the history and story of drug is old as mankind. There is a general belief that herbal drugs have no side effects, cheap and locally available so it is used as primary health care [3].

The term 'Urolithiasis' comes from the Greek word, ouron means urine and *lithos* means "stone"[4]. It is also called as nephrolithiasis, kidney stones or renal calculi. Deficiency of any one of inhibitors or excess of any one of promoters plays an important role in stone formation [5].

Pistia stratiotes is an aquatic plant, floating on lakes, streams, stagnant water ponds and lime-rich water. It is distributed in the tropical and subtropical region of Asia, Africa, and America[6]. *Pistia stratiotes* is found in ponds and streams almost allover India. *Pistia stratiotes* leaves are green color, odorless and bitter in taste. The ash of *Pistia stratiotes* is applied to the ringworm of the scalp. The Plant leaves are used in ulcers,eczema,leprosy,syphilis and piles. Juice of leaves boiled in coconut oil is applied externally in chronic skin diseases[7]. The aim of present study is to evaluate the efficacy of *Pistia stratiotes* in management of urolithiasis in sodium oxalate induced urolithiasis in wistar albino rat model.

MATERIALS AND METHODS

Plant

The plant *Pistia stratiotes* was authenticated by Dr.V.Ganesan, Associate Professor and Head, Centre for research PG Studies n Bontany, Ayya Nadar Janaki ammal College, Sivakasi, Tamil Nadu, India.

Collection of plant

Whole plant of *Pistia stratiotes* collected in the month of August 2012, from Manappakkam , Kanchipuram district , Tamilnadu , India. The whole plant were cleaned and dried under the shade to avoid degradation.

Preparation of Extract

The plant *Pistia stratiotes* was collected and it was size reduced into small pieces and shadow dried. The dried materials were coarsely powdered before maceration. After maceration the extract was distillated and crude extract was collected [8].

Animals

Wistar albino rats weighing 150-200g were used for this study. They were maintained in standard environmental conditions of temperature (25±2^{0}c), The animals were housed in clean polypropylene cages lined with husk, changed every 24 hours under a 12-hour light/ dark cycle. They were fed with standard diet and water *ad libitum*[9]. The studies were conducted in accordance with the Institutional Animal ethical committee : SBCP/2012-2013/CPCSEA/IAEC-III/04.

Experimental design

Sodium oxalate–induced urolithiatic model in rat was used to assess the activity of *Pistia stratiotes*. Rats were divided into five groups, each group containing 6 animals. Group I received only normal saline and served as normal rats. Group II which served as negative control. Group III were administered with cystone (500 mg/kg p.o) which served as positive control. Group IV and V were treated with methanolic extract of *Pistia stratiotes(MEPS)* at a dose of 250mg/kg and 500mg/kg p.o respectively[10]. Both the extracts and vehicle treatments were done for 10 days. Group II were injected with sodium oxalate (70 mg/kg, i.p.) after administration of MEPS, daily for 10 days. The urine of each rat was collected on the 10th day. Blood were collected immediately after urine collection through retro-orbital puncture under light

ether anaesthesia. The animals were sacrificed by cervical dislocation and removed the kidneys to examined for the presence of calcium oxalate crystals and stone formation by histological techniques[11].

RESULTS AND DISCUSSION

Body weight

The change in body weight of albino rats after receiving sodium oxalate (70mgkg.p.o) and respective drug treatments was shown in Table-. The Group-2 animals show significant decrease in body weight when compared to Group-1 animals. But the Group 3 to 5 animals receiving respective drug treatment shows increase in body weight when compared to Group-2.

Urine volume and pH

Administration of Sodium oxalate (70mg/kg.p.o) shows significant change in urine volume and urine pH. Administration of cystone (500mg/kg p.o.), MEPS (250 and 500mg/kg) shows significantly increase in urine volume and pH compared to sodium oxalate alone treated group. The results were shown in table-.

Urinary creatinine

Administration of sodium oxalate (70mg/kg i.p.) for 10 days significantly reduce urinary creatinine level when compared to Group-1. Treatment with MEPS (250 and 500mg/kg p.o) and cystone (500mg/kg p.o) for ten days significantly increased ($P<0.01$) the excretion of creatinine when compared to Group-2.

Urinary Urea

The group of sodium oxalate was increase when compared to Group-I. The treatment of cystone and MEPS (250 and 500 mg/kg p.o.) also significantly reduced when compared to Group-II

Uric acid and Sodium

Sodium oxalate was significantly decrease when compared to Group-I. The treatment of cystone and MEPS (250 and 500 mg/kg p.o.) significantly increased when compared to Group-II respectively.

Potassium

The Group-II (sodium oxalate) was significantly decrease when compared to Group-I. The treatment of cystone and MEPS (250 and 500 mg/kg p.o.) were significantly increased when compared to Group-II respectively

Calcium and Oxalate

Sodium oxalate(Group-II) was significantly increased when compared to Group-I. The Group-II Cystone (Standard drug) 500mg/kg.p.o was decreased and Methanolic extracts of *Pistia stratiotes* (MEPS) (250 and 500 mg/kg p.o.) also significantly decreased when compared to Group-II respectively.

Total protein

Group-II (Sodium oxalate) was significantly increased when compared to Group-I. The Group-II Cystone (Standard drug) 500mg/kg.p.o was decreased and MEPS (250 and 500 mg/kg p.o.) also significantly decreased when compared to Group-II respectively

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Effect of PS on changes body weight on sodium oxalate induced Urolithiasis.

Treatment and dose	Body weight in gram	
	Initial	Final
Normal Saline(10ml/kg.p.o)	170.8±1.53	177.5±1.11
Negative control(70mg/kg.p.o)	179.2±1.54	165.0±1.29
Cystone(500mg/kg.p.o)	178.3±1.05	180.8±1.53
MEPS(200 mg/kg.p.o)	183.3±2.11	187.5±1.12
MEPS(400 mg/kg.p.o)	165±1.82	172.5±1.11

Effect of PS on urinary output and urinary pH in sodium oxalate induced urolithiasis

Treatment	Volume of urine(ml)	pH of urine
Normal Saline(10ml/kg.p.o)	3.4±0.025	7.5±0.03
Negative control(70mg/kg.p.o)	0.64±0.005	6.36±0.03
Cystone(500mg/kg.p.o)	5.31±0.047**	7.71±0.07**
MEPS(200 mg/kg.p.o)	2.41±0.031*	6.83±0.03*
MEPS(400 mg/kg.p.o)	5.16±0.055**	7.61±0.03**

Values are mean ± SEM; n=6 in each group; Group –II was compared with Group –I. Group-III to V were compared to Group-II. The values of biochemical parameters rats were not altered significantly.

Effect of PS on urinary parameters against sodium oxalate induced Urolithiasis

Group	Urine levels			
	Creatinine (g/l)	Urea (g/l)	Uric acid (g/l)	Sodium (mEq/l)
Normal Saline(10ml/kg.p.o)	0.78±0.005	4.01±0.085	4.5±0.005	152.0±0.48
Negative control(70mg/kg.p.o)	0.23±0.004	7.88±0.31	1.63±0.003	70.13±0.51
Cystone(500mg/kg.p.o)	0.76±0.004	4.33±0.28	4.43±0.003	147.5±0.42
MEPS(200 mg/kg.p.o)	0.44±0.006*	6.10±0.22*	3.25±0.062*	139.9±0.43*
MEPS(400 mg/kg.p.o)	0.76±0.004**	5.36±0.055**	4.42±0.003**	146.8±0.34**

Values are mean ± SEM; n=6 in each group; Group –II was compared with Group –I. Group-III to V were compared to Group-II. The values of biochemical parameters rats were not altered significantly.

Effect of PS on urinary parameters against sodium oxalate induced Urolithiasis

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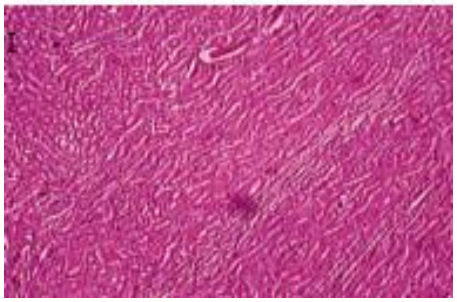
Effect of PS on urinary parameters against sodium oxalate induced Urolithiasis

Group	Urine levels			
	Potassium (mEq/l)	Calcium (mMol/l)	Oxalate (mg/dl)	Total protein (mg/dl)
Normal Saline(10ml/kg.p.o)	5.4±0.05	5.72±0.12	0.45±0.003	3.43±0.042
Negative control(70mg/kg.p.o)	2.1±0.02	15.58±0.35	1.98±0.002	8.58±0.03
Cystone(500mg/kg.p.o)	5.08±0.04	6.12±0.12	0.46±0.002	3.20±0.025
MEPS(200 mg/kg.p.o)	4.33±0.02*	7.37±0.13*	0.51±0.003*	3.83±0.03*
MEPS(400 mg/kg.p.o)	5.01±0.05**	5.95±0.26**	0.47±0.002**	3.45±0.042**

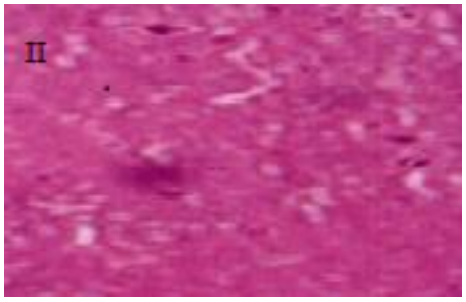
Values are mean ± SEM; n=6 in each group; Group –II was compared with Group –I. Group-III to V were compared to Group-II. The values of biochemical parameters rats were not altered significantly.

Effect of PS on Kidney histopathology of animals treating with sodium oxalate

Normal Saline

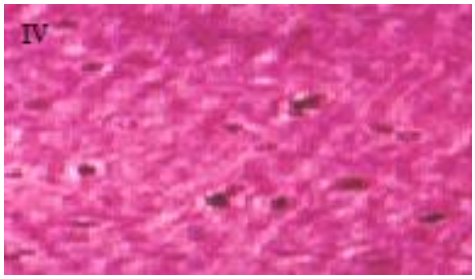


Negative control

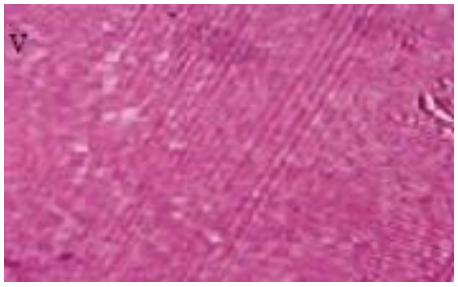


Cystone

MEPS 200



MEPS 400



Kidneys of all animals were subjected to histopathological studies. The sections of kidneys of negative control rats has shown deposition of micro crystals of sodium oxalate and crystal deposition in cortex.There was no significant tubular damage, hemorrhage and tubular congestion in the kidney sections (cortex) of the rats treated groups and the crystal deposition was significantly less when compared with disease induced animals.

CONCLUSION

The literature review revealed that there was no information available regarding the study of anti urolithiatic activity by any researcher earlier. After going through the literature review, it was clearly understood that the plant *Pistia stratiotes* has antiurolithiatic activity and the present work indicated that both concentration (200 and 400mg/kg p.o.) of methanolic extracts of *Pistia stratiotes* were possessed anti urolithiatic activity that supports the folkloric use.It was concluded that further study of methanolic extracts of *Pistia stratiotes* 400mg/kg p.o. shows highly protective action when compared to methanolic extracts of *Pistia stratiotes* 200mg/kg p.o.The studies may be extended in future by isolating the phytoconstituents by column chromatography which is responsible for anti urolithiatic activity. Further studies are in progress at department of pharmacology in our college.

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