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Anti Urolithiatic activity of Extracts of *Aerva javanica* in Rats

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Abstract:
In the indigenous system of medicine in India, the plant *Aerva javanica* commonly termed as Tella burga is claimed to be useful for various ailments, and one such use is for the treatment of renal calculi. The major purpose of this study is to investigate the potential activity of *Aerva javanica* in the treatment of renal calculi. Urolithiasis is the process of development of crystal in the urinary tract. Wistar rats were taken and they are divided into nine groups. The first group used as normal and remaining groups received ethylene glycol 0.75% by orally for 28days. Second group received ethylene glycol Group-III received cystone (750mg/kg) till 28th day. Groups IV, V, VI and VII, VIII and IX served as preventive regimen received *Aerva javanica* aqueous extract, methanolic extract and ethyl acetate extract (200mg/kg and 400mg/kg) per oral respectively from 1st day till 28th day. On the 28th day of experiment animals were housed in metabolic cages and 24 hours urine samples and serum samples were collected. The urine was subjected to microscopical study to observe the crystals. The parameters monitored in the present study are uric acid, creatinine, sodium, potassium, magnesium oxalates and calcium in urine and serum samples.
Ethylene glycol feeding resulted in hyperoxaluria as well increased renal excretion of calcium and oxalates. Treatment with aqueous, methanol and ethyleacetate extract of Aerva javanica roots significantly reduced the elevated urinary oxalate, showing a regulatory action on endogenous oxalate synthesis. The increased deposition of stone forming constituents in the serum and urine of calculogenic rats was also significantly lowered.

Keywords: Anti-Urolithiasis activity, Aerva javanica, ethylene glycol, hyperoxaluria and kidney stones.

INTRODUCTION:

Urinary stone disease is a common disorder estimated to occur in approximately 12% of the population, with a recurrence rate of 70–81% in males, and 47–60% in females. Occurrence of urolithiasis requires formation of a nidus, its retention and growth in the urinary tract which may cause obstruction of the reter1.Urolithiasis, one of the most painful ailments of the urinary tract disorder, has beset humans from centuries. Calcium oxalate (CaOx) is the primary constituent of the majority of stones formed in the urinary system of patients with urolithiasis2. About 5% of American women and 12% of men will develop a kidney stone at some time in their life, and prevalence has been rising in both sexes. Approximately 80% of stones are composed of calcium oxalate (Caox) and calcium phosphate (CaP); 10% of struvite (magnesium ammonium phosphate produced during infection with bacteria that possess the enzyme urease), 9% of uric acid (UA); and the remaining 1% are composed of cystine or ammonium acid urate or are diagnosed as drug-related stones3. Kidney stone formation or Urolithiasis is a complex process that results from a succession of several physicochemical events including supersaturating, nucleation, growth, aggregation, and retention within the kidneys. Stone formation in the kidney is one of the oldest and most wide spread diseases known to man. Urinary calculi have been found in

the tombs of Egyptian mummies dating back to 4000 BC and in the graves of North American and Indians from 1500-1000 BC. Reference to stone formation is made in the early Sanskrit documents in India between 3000 and 2000 BC5. Approximately 0.1–0.4% of the population is believed to have kidney stones every year in the USA and Europe; about 2–5% of the population in Asia, 8–15% in Europe and North America, and 20% in Saudi Arabia develop renal stones in their lifetime[1].

The world wide incidence of Urolithiasis is quite high and in spite of tremendous advances in the field of medicine there is no truly satisfactory mode of treatment for treating renal calculi. For the management of urolithiasis, combination of surgical and medical approach using percutaneous nephrolithotomy (PCNL), extracorporeal shock wave lithotripsy (ESWL) and antibiotics are employed. These treatments are relatively costly, painful and require expert hands and availability of appropriate equipments. These treatments cause undesirable side effects such as hemorrhage, hypertension, tubular necrosis and subsequent fibrosis of the kidney leading to cell injury and recurrence of renal stone formation[2]. In spite of tremendous advances in the field of medicine, there is no truly satisfactory drug for the treatment of renal calculi. Most patients still have to undergo surgery to get rid of this painful disease. So, it is worthwhile to look for an alternative for the management of Urolithiasis, and therefore phytotherapy is being sought. *Aerva javanica* is said to be useful in treating urinary calculi8. Hence the present study, an effort has been made to establish the scientific validity for the antiurolithiatic activity of *Aerva javanica* extracts using induced hyperoxaluria model in rats[3].

MATERIALS AND METHODS:

Based on ethnomedical information and literature surveys the *Aervajavanica* were collected from rural areas of Warangal dist, AP, India. The taxonomic identities of these plants were confirmed by department of botany, Kakatiya University, warangal, India[4]. A brief description of the plant and systemic position is provided. Fresh parts of four different plant species free from diseases were collected and bought to laboratory in sterile polyethylene bags and washed thoroughly 2-3 times with running tape water and then once with sterile water, shade dried for two weeks, subsequently ground into fine powder using mechanical grinder and motor driven grinding mill. The powder was used for extraction of crude extracts.

Method of extraction of crude extracts

Sequential extraction method was employed to extract to extract the plant powders using different polar solvents to non polar solvents namely aqueous, methanol and ethyl acetate. The *Aerva javanica* plant parts were subjected for continuous extraction with methanol, ethyl acetate and filtered. The filtrate was evaporated at room temperature and concentrated for dryness of extract in desiccators to remove moisture. The extractions were carried out from the plant root. In most of the cases the amount of residue extracted with methanol is higher when compared to other solvents.

Chemicals and Glassware

All chemicals and glassware used in the present investigations were obtained from the Hi-Media, SD-Fine and E-Merk India Limted (Mumbai). The glassware was thoroughly washed in chromic acid and rinsed with distilled water before use.

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Phytochemical evaluations of extracts:

Preliminary phytochemical screening of the extracts for alkaloids, carbohydrates, glycosides, saponins, phytosterols, fixed oils, fats, resins, phenols, tannins, flavonoids, proteins and aminoacids[5].

EXPERIMENTAL METHOD:

Wistar albino rats (150-200 g) were used in pharmacological and toxicological studies. The animals were purchased from the Mahaveer Enterprises,Hyderabad. The animals had free access to standard diet with water supplied *ad libitum* under strict hygienic conditions. Each experimental group had separate set of animals and care was taken to ensure that animals used for one response were not employed elsewhere. Animals were habituated to laboratory conditions for 48 hours prior to experimental protocol to minimize if any of non-specific stress. The approval of the Institutional Animal Ethical Committee (IAEC) of Jangaon institute of pharmaceutical sciences was taken prior to the experiments. All the protocols and the experiments were conducted in strict compliance according to ethical principles and guidelines provided by Committee for the Purpose o f Control and Supervision of Experiments on Animals (CPCSEA).

Acute oral toxicity:

It is the principle of the test that based on a stepwise procedure with the use of a minimum number of animals per step; sufficient information is obtained on the acute toxicity of the test substance to enable its classification. The substance is administered orally to a group of experimental animals at one of the defined doses. The substance is tested using a stepwise procedure, each step having three animals of a single sex. Absence or presence of compound related mortality of the animals dosed at one step was determining the next step.

Method:

The overnight fasted mice were divided into four groups, each group consisting of three female animals. The extracts of *Aerva javanica* root are given by gastric incubation with a syringe. After administration of the extract, the animal were observed continuously for the first two hours and at 24 hours to detect changes in behavioral responses and also for tremors, convulsion, salivation, diarrhoea, lethargy, sleep, coma and also were monitored up to 14days for the toxic symptoms and mortality.

ASSESSMENT OF ANTI-UROLITHIATIC ACTIVITY OF *AERVA JAVANICA*

Ethylene glycol induced hyperoxaluria model was used to assess the antilithiatic activity in albino rats. The study was designed to find out the effect of alcoholic and aqueous extracts of *Aerva javanica* usage against Ethylene glycol induced urolithiasis.

Preparation and administration of doses

All the doses were prepared in distilled water using 10% tween 80 as suspending agent. In all cases the concentrations were prepared in 1 ml/100 mg of b.w. The test substances were administered orally in a single do se using a gastric intubation tube after fasting for 3 to 4 hours.

Evaluation of antiurolithiatic activity:

Animals were divided into 9 groups containing six in each and kept in metabolic cages individually for entire duration of the experiment. All animals had free access to regular rat chow and drinking water *ad libitum*. Renal calculi were induced in group II to IX by Ethylene glycol (0.75%) in drinking water *ad libitum* for 28 days. Group III has treated with the cystone as a standard. Group IV to IX

were treated with aqueous, methanol and ethyl acetate extracts of *Aerva javanica* roots starting from 1 day to 28 day. All rats were housed in metabolic cages individually for entire duration of the experiment. On 28th day, 24 hours after the treatment, all the animal were hydrated with Saline(20ml/kg) urine samples were collected from all the animals and serum samples from each rat were collected. pH of urine were checked by using narrow ranges pH (BDH) paper and urine volumes were noted. A drop of concentrated hydrochloric acid was added to the urine before being stored at 4oC. The presence of calcium oxalate crystals were observed by urine sediment using microscopic techniques.

Protocol for Antiurolithiatic activity

Group I Normal control

Group II Ethylene glycol

Group III Ethylene glycol + cystone

Group IV Ethylene glycol +Aqueous extract of *Aerva javanica*.(200mg/kg b.w.,p.o.)

Group V Ethylene glycol +Aqueous extract of *Aerva javanica* (400mg/kgb.w.,p.o.)

Group VI Ethylene glycol + methanol extract of *Aerva javanica* (200mg/kg b.w.,p.o.)

Group VII Ethylene glycol + methanol extract of *Aerva javanica* (400mg/kg b.w.,p.o.)

Group VIII Ethylene glycol + ethyl acetate extract of *Aerva javanica* (200mg/kg b.w.,p.o.)

Group IX Ethylene glycol + ethyl acetate extract of *Aerva javanica* (400mg/kg b.w.,p.o.).

Collection and analysis of urine:

All rats were housed in metabolic cages individually. On 28th day, 6 hours after the treatment, all animal were hydrated with Saline (20ml/kg). Urine samples were collected from all the animals. Magnesium, uric acid, creatinine, sodium, potassium, calcium and oxalates content were determined. Urine was centrifuged to pool the crystals and observed under light electron microscope at 5X or 10X.

Serum analysis:

After the experimental period, the animals were sacrificed by cervical decapitation under anesthetic conditions and blood was collected from the retro orbital. Serum was separated by centrifugation at 10,000 rpm for 10 min and analyzed for magnesium, uric acid, creatinine, sodium, potassium, calcium and oxalates.

MICROSCOPIC STUDIES:

Microscopic examination should be performed on centrifuged sample.

Sediment preparation and use of microscope:

- Mix the specimen and then place approximately 10-15 ml of urine into a centrifuge tube.
- Centrifuge at 2000 rpm for about 5minute.
- Pour off the supernatant fluid & resuspeneded the sediment in the urine that drains back down from the side of the tube.
- Mix the sediment and place a drop of sediment on a clean slide.
- Cover coverslip and examine immediately.

In centrifuged urine sediment, *looks for* cellular elements (RBCs, WBCs, Epithelial cells), cast, crystals, miscellaneous structure (bacteria, yeast, Mucus thread) and possible parasites, etc. Those *noted* element *reported* according to type as well as by giving an estimation of their number. Cast reported under low power field (LPF=10X), while RBCS and WBCs counted under high power field (HPF=40X).

- The background matrix of the cast may be hyaline or granular in nature.
- Often, they are seen in urines in which free lipid droplets are present as well.

Crystals:

- Crystals are usually not found in fresh urine but appear after the urine stand for a while.
- Many of them have a little clinical significant.
- Since crystal formation tend to be pH dependent, it is helpful to be aware of the pH of the urine when performing microscopic examination.

Calcium Oxalate:

- Resembling an envelope or whole peanut "very refractile"
- It can be present in normal urine.
- Increased amount of Calcium Oxalate suggest the possibility of oxalate calculi.
- Pathological conditions include: diabetes mellitus, liver disease and severe Chronic renal disease.

Amorphous urates:

- Yellow granular appearance, grouped in compact cluster.
- Has no clinical significant.

PRELIMINARY PHYTOCHEMICAL SCREENING:

The extracts of drug were analyzed for the presence of various constituents. The result of this preliminary phytochemical examination there is presence of flavonoids, glycosides, terpenoids, steroids and cardiac glycoside in aqueous, methanol and ethylacetate extracts.

PHARMACOLOGICAL ACTIVITY

Acute oral toxicity:

Acute oral toxicity was carried out according to OECD guidelines. AEAJ, MEAJ and EEAJ were safe up to 2000mg/kg and were lethal at 5000mg/kg dose.

Anti-urolithiatic activity of roots of *Aerva javanica*

Urinary volume and pH:

S. No	Group	Volume of urine	Urinary pH
1	Normal urine	12.20±0.59	7.94±0.20
2	Ethylene glycol	6.047±0.48	6.110±0.26
3	Cystone	12.11±0.64***	7.78±0.20***
4	AEAJ 200mg/kg	11.61±0.68***	7.9±0.18***
5	AEAJ 400mg/kg	12.32±1.13***	8.15±0.19***
6	MEAJ 200mg/kg	10.97±0.77**	7.32±0.23**
7	MEAJ 400mg/kg	11.67±1.01***	7.98±0.24***
8	EEAJ 200mg/kg	9.88±0.99*	7.15±0.28*
9	EEAJ 400mg/kg	10.3±1.07**	7.26±0.24**

Values are Mean ± SEM. (n=6) significance values are ***p<0.001,
**p<0.01 and *p<0.05. Control group VS all groups.

Effect of *Aerva javanica* root extracts on serum constituents against Ethylene glycol induced urolithiasis

Groups	Sodium mEq/ml	Potassium mEq/ml	Calcium mg/dl	Creatinine mEq/dl	Magnesium mEq/l	Uric acid Mg/dl	Oxalates Mg/dl
Normal(saline)	121.0±0.89	4.13±0.19	4.40±0.21	0.43±0.08	1.16±0.08	1.15±0.12	1.23±0.03
Ethylene glycol	160±2.06	6.01±0.06	0.88±0.11	1.28±0.12	1.76±0.05	2.2±0.26	2.64±0.20
Cystone	121.7±0.57***	3.16±0.09***	4.58±0.20***	0.44±0.04***	1.22±0.06***	0.89±0.13***	1.52±0.06***
AEAJ 200mg/kg	155.2±0.41**	3.78±0.07***	1.81±0.13**	0.62±0.12**	1.47±0.01**	1.30±0.13**	1.93±0.10**
AEAJ 400mg/kg	143.4±0.82***	3.15±0.10***	4.38±0.20***	0.44±0.06***	1.29±0.09***	0.99±0.77***	1.64±0.15***
MEAJ 200mg/kg	156.0±0.64**	5.56±0.11*	1.80±0.19**	0.71±0.09*	1.45±0.05**	1.29±0.20**	2.01±0.19*
MEAJ 400mg/kg	155.2±0.39***	5.4±0.10**	3.55±0.16***	0.62±0.05**	1.33±0.02***	1.09±0.13***	1.79±0.11***
EEAJ 200mg/kg	158.3±0.26	5.67±0.14	1.51±0.15	0.84±0.21	1.55±0.05	1.67±0.15	2.14±0.17
EEAJ 400mg/kg	156.0±0.36*	5.56±0.11**	1.51±0.41	0.86±0.24	1.55±0.04	1.69±0.07	2.10±0.12

Values are mean ± S.E.M.(n=6);significance values are ***p<0.001, **p<0.01 and *p<0.05 control group vs all groups.

Effect of *Aerva javanica* root extracts on urine parameters against ethylene glycol induced urolithiasis

Groups	Sodium mEq/24hrs	Potassium mEq/24hrs	Calcium Mg/24hrs	Creatinine Mg/24hrs	Magnesium mEq/24hrs	Uric acid Mg/24hrs	Oxalates Mg/24hrs
Normal (saline)	133.4±1.45	45.23±0.91	5.83±0.65	1.71±0.13	5.21±0.18	1.00±0.14	1.23±0.03
Ethylene glycol (control)	80.03±1.17	35.01±0.91	8.86±0.36	0.45±0.06	1.74±0.09	2.4±0.16	9.00±0.09
Cystone	136.3±1.31***	49.64±0.90***	6.5±0.23***	1.62±0.08***	5.32±0.34**	0.93±0.14***	6.02±0.12***
AEAJ 200mg/kg	88.08±2.55**	40.0±1.43*	7.15±0.19**	1.22±0.18**	3.17±0.29***	1.16±0.10**	7.76±0.10**
AEAJ 400mg/kg	133.1±0.82***	46.13±0.77***	5.0±0.16***	1.64±0.07***	5.18±0.42***	1.06±0.11***	6.17±0.20***
MEAJ 200mg/kg	87.79±0.835**	39.9±1.46*	7.48±0.37*	1.00±0.16*	2.88±0.20*	1.55±0.13**	7.97±0.50*
MEAJ 400mg/kg	106.0±2.58***	41.22±1.29**	6.90±0.16**	1.21±0.15**	3.10±0.28**	1.40±0.28***	6.94±0.34***
EEAJ 200mg/kg	85.95±0.879	36.85±1.00	8.00±0.14	0.93±0.13	2.75±0.21	1.78±0.09	8.38±0.16
EEAJ 400mg/kg	87.02±0.894*	37.93±1.74*	7.55±0.37*	0.93±0.13	2.88±0.20*	1.81±0.18	8.40±0.10

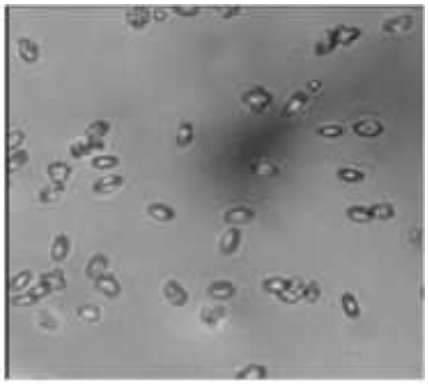
Values are mean ± S.E.M.(n=6);significance values are ***p<0.001, **p<0.01 and *p<0.05 control group vs all groups.

Effect of *Aerva javanica* root extracts on urine parameters against ethylene glycol induced urolithiasis

Groups	Sodium mEq/24hrs	Potassium mEq/24hrs	Calcium Mg/24hrs	Creatinine Mg/24hrs	Magnesium mEq/24hrs	Uric acid Mg/24hrs	Oxalates Mg/24hrs
Normal (saline)	133.4±1.45	45.23±0.91	5.83±0.65	1.71±0.13	5.21±0.18	1.00±0.14	1.23±0.03
Ethylene glycol (control)	80.03±1.17	35.01±0.91	8.86±0.36	0.45±0.06	1.74±0.09	2.4±0.16	9.00±0.09
Cystone	136.3±1.31***	49.64±0.90***	6.5±0.23***	1.62±0.08***	5.32±0.34**	0.93±0.14***	6.02±0.12***
AEAJ 200mg/kg	88.08±2.55**	40.0±1.43*	7.15±0.19**	1.22±0.18**	3.17±0.29***	1.16±0.10**	7.76±0.10**
AEAJ 400mg/kg	133.1±0.82***	46.13±0.77***	5.0±0.16***	1.64±0.07***	5.18±0.42***	1.06±0.11***	6.17±0.20***
MEAJ 200mg/kg	87.79±0.835**	39.9±1.46*	7.48±0.37*	1.00±0.16*	2.88±0.20*	1.55±0.13**	7.97±0.50*
MEAJ 400mg/kg	106.0±2.58***	41.22±1.29**	6.90±0.16**	1.21±0.15**	3.10±0.28**	1.40±0.28***	6.94±0.34***
EEAJ 200mg/kg	85.95±0.879	36.85±1.00	8.00±0.14	0.93±0.13	2.75±0.21	1.78±0.09	8.38±0.16
EEAJ 400mg/kg	87.02±0.894*	37.93±1.74*	7.55±0.37*	0.93±0.13	2.88±0.20*	1.81±0.18	8.40±0.10

Values are mean± S.E.M.(n=6);significance values are ***p<0.001,**p<0.01 and *p<0.05. control group vs all groups

MICROSCOPICAL STUDIES



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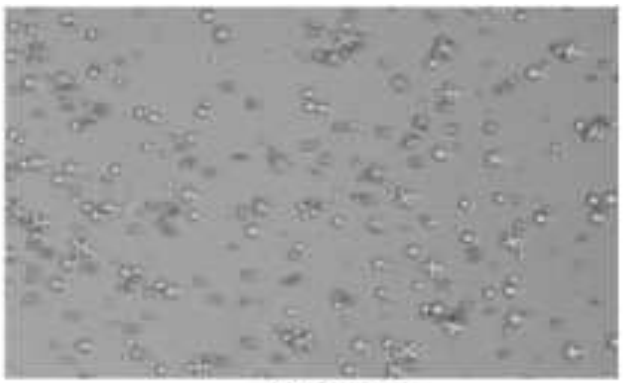

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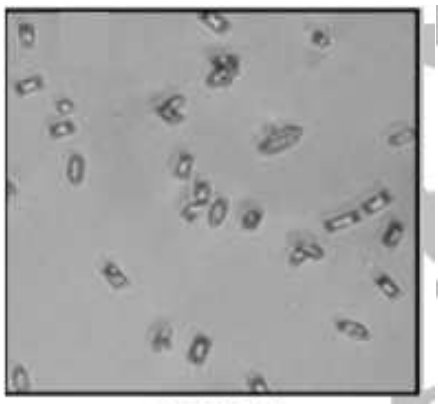

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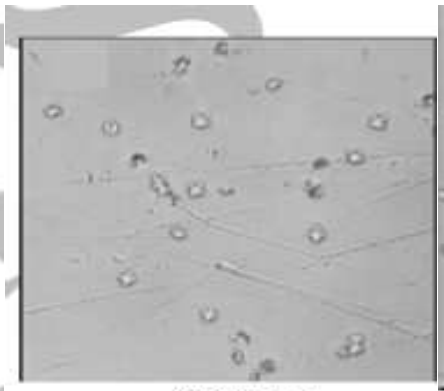

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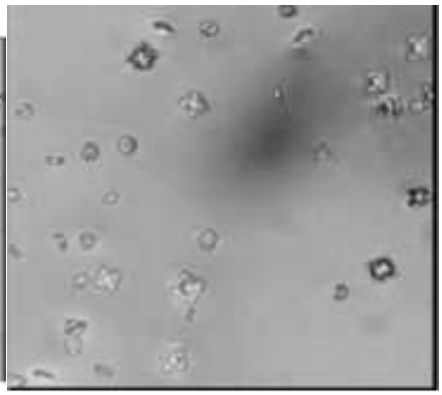

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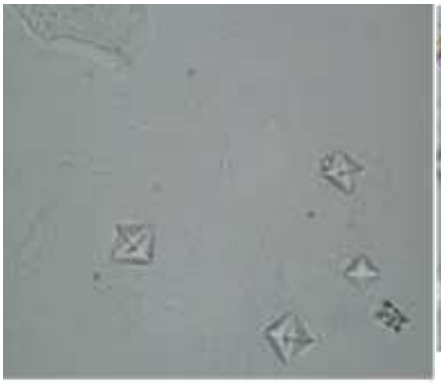

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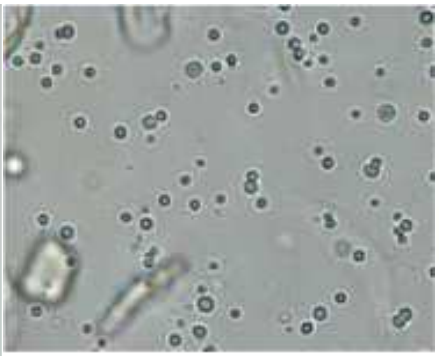

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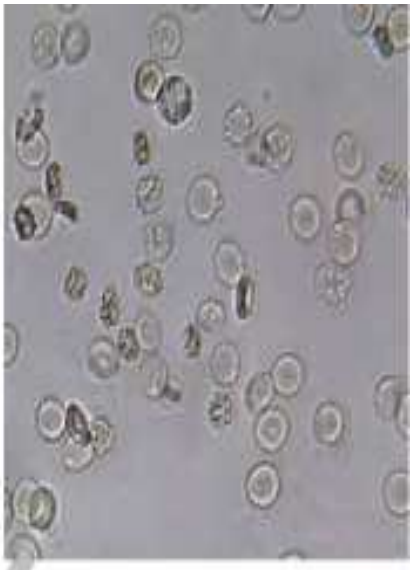

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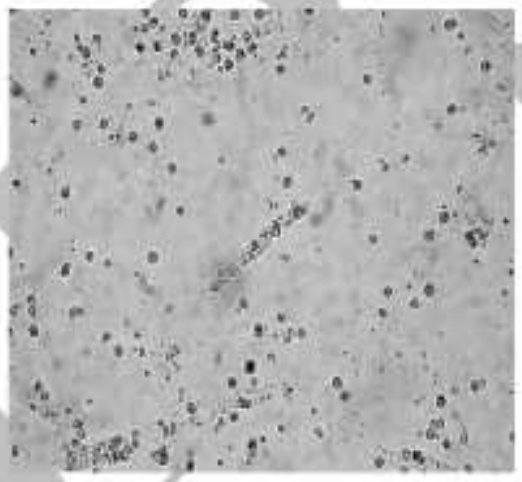

Group IX

Discussion:

As traditional medicines are usually taken by the oral route, same route of administration was used for evaluation of protective effect of the *Aerva javanica*against ethylene glycol induced urolithiasis in rats. The discoveries of the clinical roles of these herbal remedies have made important contributions to the treatment of urinary stone disease as an alternative or adjunct therapy. Kidney stones develop as a result of a complicated interaction of biological events that are most likely triggered by genetic susceptibility coupled to dietary factors and lifestyle. In the present study, male rats were selected to induce Urolithiasis because the urinary system of male rats resembles that of humans and also earlier studies have shown that the amount of stone deposition in female rats was significantly less. In the present study, chronic administration of 0.75 % (v/v) ethylene glycol aqueous solution to male Wistar rats resulted in hyperoxaluria. Oxalate and calcium excretion were grossly increased in calculi-induced animals. Since it is accepted that hyperoxaluria is a far more significant risk factor in the pathogenesis of renal stones than hypercalciuria, the changes in urinary oxalate levels are relatively much more important than those of calcium. Increased urinary calcium is a factor favoring the nucleation and precipitation of calcium oxalate or apatite (calcium phosphate) from urine and subsequent crystal growth. However, supplementation with AEAJ, MEAJ and EEAJ of *Aerva javanica* significantly lowered the elevated levels of oxalate as well as calcium excretion in urine. Normal urine contain s many inorganic and organic inhibitors of crystallization, magnesium is one such well-known inhibitors. Low levels of magnesium are also encountered in stone formers as well as in stone-forming rats. The magnesium levels return to normal on drug treatment with *Aerva javanica*, but the preventive regimen study increases the magnesium level more comparing to the curative regimen study. The increase in urinary uric acid excretion was observed in urolithiatic rats. Increased excretion of uric acid has been reported in stone formers and hyperoxaluric rats. Uric acid interferes with calcium oxalate solubility and it binds and reduces the inhibitory activity of glycosaminoglycans.

The predominance of uric acid crystals in calcium oxalate stones and the observation that uric acid binding proteins are capable of binding to calcium oxalate and modulate its crystallization also suggests its primary r ole in stone formation. Treatment of *Aerva javanica* lowered the excretion of uric acid and reduces the risk of stone formation. The present study examined the effect of various extracts, doses and studies (PR) of *Aerva javanica* roots on creatinine clearance in urine. Inducing agent showing a decrease in creatinine level which increased by cumin this indicates the anti lithiatic action of *Aerva javanica.* In urolithiasis, the glomerular filtration rate (GFR) decreases due to the obstruction to the outflow of urine by stones in urinary system. Due to this, the waste products, particularly nitrogenous substances such as creatinine and uric acid get accumulated in blood. In the present study, the positive control calculi-induced rats were found to have marked renal damage, consistent with the elevated serum levels of creatinine and uric acid. However, the curative and prophylactic treatment with PR AEAJ, MEAJ and EEAJ inhibited these changes that would otherwise promote new stone formation in the urinary system. In rats treated with *Aerva*

javanica, we attribute the lower serum creatinine and uric acid levels to an enhanced GFR. The mechanism of antilithiatic activity of aqueous extract and alcoholic extract may involve the inhibition of oxalate induced toxic manifestations and free radical production along with enhancement of the body defense system. Drug treated group showing cytoprotection due to its effect on prevention of deposition or aggregation of calcium oxalate in tubules so the mechanical disruption of epithelium is less or protection against free radicals rearrangements. The alterations in membrane lipid and protein resulting from peroxidation can lead to increased permeability of calcium, resulting in a loss of enzyme activity. Decreased Ca2+ ATPase activity was noted in the kidneys of patients with nephrolithiasis. Up on treatment 13 with roots of *Aerva javanica* , there was an elevation in the levels of calcium in preventive group when compared to control and this effect was dose dependent. In this study, 28 days administration of 0.75% (v/v) ethylene glycol induced nephrotoxicities. These toxicities were characterized by marked elevation of blood magnesium and oxalates are remarkable decrease to normal value by treating with aqueous, methanol and ethyl acetate extract of *Aerva javanica*. In case of urolithiasis, elimination of the normal electrolytes like sodium and potassium will be reduced due to the deposition of the crystals in distal tubule of the kidney where those parameters will get excreted. Hence due to the reduced elimination serum level of those normal electrolytes will be elevated. In our present investigation reveals that PR of AEAJ, MEAJ and EEAJ increases the electrolytes elimination and hence reduced their blood level to normal. Microscopic examination using polarized light of urine sediment derived from nephrolithiatic rats showed intra tubular and interstitial crystal deposits, consistent with the findings of others. In the present investigation microscopically evaluation showed the maximum prevention of crystals deposition at the preventive study which may be due to the active compound which is present in aqueous , methanol and ethyl acetate extracts of *Aerva javanica roots* a high antioxidant capacity may be due to the presence of important phytoconstituents like, phenolic compounds, steroids and flavonoids may be prevents the calcium oxalate crystal deposition in the kidney by preventing hyperoxaluria- induced peroxidative damage to the renal tubular membrane surface which in turn can prevent calcium oxalate crystal attachment and subsequent development of kidney stones.

Conclusion

The presented data indicate that administration of the ethyl acetate, methanolic and aqueous extracts of root of *Aerva javanica* to rats with ethylene glycol induced lithiasis reduced and prevented the growth of urinary stone, supporting folk information regarding antiurolithiatic activity of the plant. Result indicates aqueous and alcoholic extract prevents renal stones better than ethyl acetate extracts. The mechanism underlying this effect may be due to increased diuresis and lowering the urinary concentrations of stone forming constituents. Further study is in progress for identification of the active constituents of the plant.

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Summary:

The anti-urolithiatic effect of aqueous, Methanolic and ethyl acetate extract of *Aerva javanica* have been investigated in the present study. Preliminary Phytochemical investigation was carried out for the extracts to identify various Phytochemical constituents present in them. It was found that the ethyl acetate extract of *Aerva javanica* (EEAJ) contains carbohydrates, fixed oil, fats phytosterols and saponins. The methanolic extract of *Aerva javanica* (MEAJ) contains phytosterols, glycosides, saponins, carbohydrates, proteins, amino acid, flavonoids, tannins and phenolic compounds. The aqueous extract of *Aerva javanica*(AEAJ) contains carbohydrates, proteins, amino acid, phytosterols , glycosides, saponins, flavonoids, tannins and phenolic compounds. Acute oral toxicity studies were performed to find out the test dose according to the Organization of Economic Co-operation and Development (OECD) guidelines-423 and aqueous extract extract of *Aerva javanica* AEAJ, MEAJ and EEAJ were found to be safe up to 2000 mg/kg b.w. The anti-urolithiatic activity was observed experimentally induced urolithiasis i.e. ethylene glycol induced urolithiasis in rats. Ethyl acetate, methanolic and aqueous extract of root of *Aerva javanica* has showed significant anti-urolithiatic activity by maintaining the normal serum levels of Magnesium, Uric acid, Creatinine, Sodium, potassium, Calcium and Oxalates this effect was due to the effect of the extracts on the function of kidney. Microscopical studies of the urine Samples confirmed the anti-urolithiatic activity of the AEAJ, MEAJ and EEAJ. It was observed that AEAJ, MEAJ having more potent anti-urolithiatic activity in ethylene glycol induced urolithiasis rat model while EEAJ show less potent antiurolithiatic activity.

Bibliography:

1) Viel TA, Domingos CD, Monteiro APDS, Lima-Landman MTR, Lapa AJ, Souccar C. Evaluation of the antiurolithiatic activity of the extract of *Costus spiiralis Roscoe* in rats. *J Ethnopharmacol* 1999; 66:193-8.
2) Kaur T, Bijarnia RK, Singla SK, Tandon C. In vivo efficacy of *Trachysper mumammi* anticalcifying protein in urolithiatic rat model. *J Ethnopharmacol* 2009; 126: 459-62.
3) Coe FL, Evan A, Worcester E. Kidney stone disease. *J Clin invest.*2005; 115: 2598-2608.
4) Takhtajan A. (2009). Flowering Plants Springer Verlag.
5) Brain K R and Turner T D. (1975). The practical evaluation of phytopharmaceuticals. *2nd edn. Bristol: Wright Sciencetechnica.*, pp. 81– 82.
6) Bouanani S ,Henchiri C , Migianu-Griffoni E, Aouf N , Lecouvey M. Pharmacological and toxicological effects of *Paronychia argentea* in experimental calcium oxalate nephrolithiasis in rats. J Ethnopharmacol 2010; 129: 38–45.
7) Doddola S, Pasupulathi H, Koganti B, Prasad KVSRG. Evaluation of *Sesbania grandiflora* for antiurolithiatic and antioxidant properties. *J Nat Med* 2008:62:300-7.
8) Bahuguna Y, Rawat MSM, Juyal V, Gupta V. Antilithiatic effect of flowers of *jasminum auriculatum* vahl. Int J Green Pharmacy 2009; 07: 155-8.
9) Betanabhatla KS, Christina AJM, Sundar BS, Selvakumar S, Saravanan KS. Antilithiatic activity of *Hibiscus sabdariffa* linn. On ethylene glycol-induced lithiasis in rats. Natural product radiance 2009; 8: 43-47.
10) Divakar K, Pawar AT, Chandrasekhar SB, Dighe SB, Divakar G. Protective effect of the hydro-alcoholic extract of *Rubia cordifolia* roots against ethylene glycol induced urolithiasis in

Covered in Scopus & Embase, Elsevier **Int. J. Drug Dev. & Res., October-December 2014, 6 (4): 35-45**

rats. Food and Chemical Toxicology 2010; 48: 1013– 1018.

11) Srinivas Reddy K and Reddy V M. (2009). Antimicrobial studies on the leaves of *Aerva Javanica. J. Pharm. Res.,* 2(7): 1259-1261.

12) Srinivas Reddy and Reddy VM. (2009). Pharmacological evaluation of methanolic extract of *Aerva javanica* leaves for central Nervous system depressant activity. *Ther Pharmacist.,* 4(1):10-12.

13) OECD 2001-gudeline on acute oral toxicity (AOT) Environmental health and safety monograph series on testing and adjustment No.423.

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