

Evaluation of Anti-Urolithiatic Activity of Selected Medicinal Plants and Its Poly Herbal Formulation in Experimentally Induced Urolithic Rats

Subhranshu Panda¹, E. Nikhil Chakravarthy², G. Tharun Kumar³

¹School of Pharmaceutical Sciences, Jaipur National University, Jaipur, India.

²School of Pharmaceutical Sciences, Jaipur National University, Jaipur, Rajasthan-302017, India.

³Osmania University, Hyderabad-500007, India.

ABSTRACT

Nelumbo nucifera, *Anogeissus latifolia*, and *Leucas aspera* (Family: Nymphaeaceae, Combretaceae, and Labiatae) have been selected for nephroprotective studies using the methanolic extract of the plants (NNE, ALE, & LAE) in rats. Kidney stones form when minerals and salts in the urine accumulate and crystallize in the kidneys or urinary tract. The Methanolic extract of NNE, ALE, LAE & and PHF of (500mg/kg, 500mg/kg & and 200mg/kg) exhibited significant urolithic action in ethylene glycol induction in rats. Cystone can be used as a standard drug and compared with the plant extracts, considering $P < 0.001$ as a significant value. In the procedure, renal analysis parameters were decreased and inhibited to a lower extent. Inducing of NNE, ALE, LAE & and PHF significantly prevents the growth of urinary stones. The possible mechanism is mediated by the diuretic, antioxidant, and anti-inflammatory effects of the plants. The results concluded that MNNE, ALE, & LAE, and PHF possessed significant anti-urolithiatic activity. These assertions show that the extracts have the pharmacological actions in the tradition of Indian medications.

Keywords: Anti-Urolithic, Urea, Nitrogen, Nephron-Protective, Diuretics

1. INTRODUCTION

Certain medical conditions, such as urinary tract infections, hyperparathyroidism, and Crohn's disease, can increase the risk of developing kidney stones¹. Certain medications, such as alpha-blockers and calcium channel blockers, can help relax the muscles in the urinary tract, making it easier for the stones to pass. Extracorporeal Shock Wave Lithotripsy: It is a process that cuts the waves of shock in which the stones can turn in the form of small residues. Uteroscopy is a better criterion in renal diagnosis. Percutaneous Nephrolithotomy (PCNL): It is suggested for the expulsion of bigger stone complexes². In rare cases, open surgery may be necessary to remove very large or complex stones. *Anogeissus latifolia*, *Leucas aspera* Linn, *Leucas aspera* & *Nelumbo nucifera*, had no acute oral toxicity when administered to Wistar rats. In the present study, it was found that the higher dose of the individual plant extracts had significant anti-urolithic activity. The polyherbal Formulation of these three plants also showed a significant decrease in urolithiasis. Saponins and quinoline alkaloids might be the cause of the therapeutic

effect. Further investigation will help to establish the phytochemical screening action that shows the desired inhibitory effects.

Nelumbo nucifera belonging to the Nymphaeaceae family includes treatment in Ayurveda this plant is used as a diuretic and anti-helminthic and in the treatment of strangury, vomiting, leprosy, skin diseases, and nervous exhaustion. In popular medicine, it is used in the treatment of tissue inflammation, cancer, skin diseases, leprosy, and as a poison antidote.

Anogeissus latifolia belongs to the family Nymphaeaceae shows treatment of skin injuries, particularly in rats.

Leucas aspera has the antifungal action of extracts containing chloroform and ether of *L. aspera* against *Microsporum gypseum* & *Trichophyton* was discovered in vitro. About 5mg/mL was determined to be the lowest inhibiting concentration. *Leucas aspera* also known as Thumbai (in Tamil), spread in various regions across the world³. Both fungicidal & fungi-static properties were found in *Leucas aspera*. Anti-oxidant activity and Prostaglandin inhibitory effects Prostaglandin (PG) inhibiting and antioxidant properties of *Leucas aspera* were investigated. The alcoholic (ethanol namely) extract was tested for anti-oxidant, anti-nociceptive, and cytotoxic activity with free-radicals, writhing inhibition mediated by acetic acid, and brine shrimp lethality bioassay, respectively. At amounts of 500 and 250 mg/kg, an ethanolic concentrate of *L. aspera* root significantly reduced acetic acid-induced writhing in mice. With an 8g/ml of IC₅₀, the extract demonstrated free radical scavenging behavior. To brine shrimp, the extract was extremely lethal.

2. MATERIALS AND METHODS

2.1 Collection of Plant material and Preparation of Extract

Nelumbo nucifera L., rhizome, *Anogeissus latifolia* - Leaves, and *Leucas aspera* Leaves were collected from Sri Venkateswara University, Tirupati, 517502, Andhra Pradesh, India. It was then certified by Dr. K. Madhava Chetty, Plant Taxonomist, from the district of Tirupati, AP bearing serial plant numbers 1078, 1080, and 1088 were collected and air-dried for seven days at room temperature and under shady conditions. The Dried plant materials are ground and sieved. The plant's powders are subjected to Soxhlet extraction using methanol as solvent packed into a soxhlet column and extracted with 70% v/v methanol in water at 65–70°C. The concentrated extract is used for the screening of possible phytochemicals⁴. The obtained methanolic extract of *NNE*, *ALE*, and *LAE* was subjected to evaporate at the temperature scale of 45°C and proceeded for further studies. The extract was preserved in an air -tight conditions.

2.2 CHEMICAL AGENTS AND APPARATUS REQUIRED

Ethylene Glycol-AR, (Merck, India), and Cystone (Himalaya Company Limited, India) are selected for the practical studies. If needed selection of the required chemicals and reagents were included with an analytical grades. Auto analyser – Semi followed by UV-spectrophotometer (UV-1700 Pharma-spec, SHIMADZU. CAT. No. 208 24201-34) etc and other apparatus can be selected on the basis for the required necessities of experimental studies⁵.

2.3 ANIMALS

Wistar rats (of both sex) weighing 150-200g and Albino mice (male) were acquired. Animals were subjected to acclimatization for laboratory conditions. Proper, hygienic, environmental conditions like temperature, relative humidity, *ad libitum* – standard for feeding were provided according to quarantine conditions. All the work carried out on the animals was in accordance with the CPCSEA guidelines and the Research protocols have been approved by the IAEC, JLS, and the Approval. No. was CPCSEA/IAEC/JLS/18/07/22/029 of Jeeva Life Sciences animal house, Medchal, Malkajgiri, Telangana. Experimental research studies were conducted and performed according to CPCSEA & IAEC rules and regulations.

A. Ethylene Glycol Induced Urolithiasis Rat Model

To develop CaOx crystals, all rats were fed with a commercially pelleted diet with a drinking water containing 0.75% EG for 28 days.

B. Calcium Oxalate Induced Urolithiasis Rat Model

42 Wistar rats weighing (150-200mg) have been divided into the following groups, by considering six animals. Considering n=6 in the following manner of Group I as a normal control carrier with normal saline solution, Group II as a toxic control with CaOx 70 mg/kg, i.p., Group III with Cystone (750 mg/kg, p.o.) as a standard drug control, with CaOx 70 mg/kg, i.p. CaOx 70 mg/kg, i.p., for Groups Groups IV, V received, CaOx with 70 mg/kg, i.p. 250 & 500mg/kg, p.o., NNE group VI, VII received 250mg/kg., 500mg/kg., of *ALE* extract and 0.75% Ethylene glycol & High Dose, group VIII, IX received low and high doses followed by 0.75% Ethylene glycol & low Dose, group X received *PHF* with 0.75% Ethylene glycol subjected with a low and high dose respectively⁶.

C. Preliminary Phytochemical Analysis

Preliminary phytochemical analysis of *NNE*, *ALE*, *LAE* along with performed. Certain phytochemical screening tests were done in order to investigate the presence of the different chemical constituents by either Ochuko & Obadoni¹⁶ or AlCl₃ method.

D. Anti-urolithiatic activity of methanolic extract of *NNE*, *ALE* and *LAE*

Low and High Doses were selected for evaluation of anti-urolithiatic activity for *NNE*, *ALE*, *LAE* along with *PHF* in this study.

Acute-toxicity studies

Toxicity studies were conducted according to Organization for Economic Cooperation and Development rules and regulations.

3. URINE SAMPLE PARAMETERS ANALYSIS

Urine samples were collected on 28th day, Phenobarbital was employed to obtain blood samples. Urinary serum parameters were analyzed and were centrifuged at 10000g for 10 minutes⁷.

4. KIDNEY'S HISTOLOGICAL STUDIES

Isolation of the kidneys were done and maintained at a temperature of 80°C along with their dried weight. On 28th day, superfluous tissues were removed. GSH, LDH, LPO were estimated with the units measured in nmol/mg of protein, nmole/min/mL, nmole/mL and the suspension was centrifuged. Coagulation of paraffin and egg albumin is the process that is used to adhere tissue to a slide. This is because eosin is an acid stain, which means that it gives the cytoplasm of pink colour. All of the cell's acidic components appear blue when stained with haematoxylin, a basic stain. The kidney with calcium oxalate deposits are elucidated with a microscopical studies.

5.1 STATISTICAL ANALYSIS

All the results were estimated according to ANOVA with a Tukey's multiple test at which Mean $\pm$ SEM with $P < 0.05$ taken in to the prime concert of the significance.

6. RESULTS

Preliminary phytochemical Screening analysis

In this studies, carbohydrates, flavonoids, tannins, terpenoids, cardiac glycosides were screened according to the pharmacognostic studies⁸.

Table 01: Methanolic extracts of NNE, ALE, LAE and PHF with Eth.G. (0.75% v/v) Induction on Renal parameters

Groups	Treatment	Descriptive Tests				
		Creatinine (mg/dL)	Calcium (mg/dL)	Phosphorus (mg/dL)	Uric acid (mg/dL)	ALP (IU/L)
Normal	Saline	0.65 $\pm$ 0.007	3.62 $\pm$ 0.006	2.94 $\pm$ 0.014	1.77 $\pm$ 0.009	75.67 $\pm$ 0.47
Control	Ethylene glycol (0.75%v/v)	0.96 $\pm$ 0.007	8.78 $\pm$ 0.005	5.72 $\pm$ 0.046	3.75 $\pm$ 0.009	96.08 $\pm$ 0.58
Standard control	Cystone (750 mg/kg, p.o.)	0.75 $\pm$ 0.005	4.23 $\pm$ 0.27	4.57 $\pm$ 0.064	2.18 $\pm$ 0.035	78.76 $\pm$ 0.77
Test Group - I	NNE (250mg/kg, p.o.)	0.75 $\pm$ 0.006	5.83 $\pm$ 0.066	5.23 $\pm$ 0.05	2.29 $\pm$ 0.049	79.78 $\pm$ 0.306
Test Group - II	NNE (500mg/kg, p.o.)	0.64 $\pm$ 0.005	4.34 $\pm$ 0.082	4.25 $\pm$ 0.028	2.11 $\pm$ 0.01	67.19 $\pm$ 0.305
Test Group - III	ALE (250mg/kg, p.o.)	0.86 $\pm$ 0.006	6.64 $\pm$ 0.069	4.95 $\pm$ 0.087	2.88 $\pm$ 0.014	83.59 $\pm$ 0.41
Test Group - IV	ALE 500mg	1.68 $\pm$ 1.005	4.76 $\pm$ 1.091	3.76 $\pm$ 1.02	2.76 $\pm$ 0.008	76.33 $\pm$ 0.33
Test Group - V	LAE (250mg/kg, p.o.)	1.162 $\pm$ 0.009	6.96 $\pm$ 0.014	4.78 $\pm$ 0.007	2.93 $\pm$ 0.008	79.81 $\pm$ 0.6
Test Group -	LAE (500mg/kg,					

VI	p.o.)	0.675±0.007	5.68±0.008	4.16±0.014	2.185±0.01	65.13±0.54
Test Group - VII	PHF (250mg/kg, p.o.)	1.597±0.003	6.57±0.013	4.77±0.009	4.0±0.02	84.63±0.41
Test Group – VIII	PHF 500mg	1.94±1.005	7.48±1.009	4.45±1.017	4.965±1.03	64.31±0.6

NNE, ALE, LAE and PHF: Methanolic *Nelumbo nucifera*, *Anogeissus latifolia*, *Leucas aspera* and PHF extracts at which *P<0.05 & 0.01 is taken for Tukeys test of significance.

Kidney failure is indicated by an elevated creatinine level. The creatinine level in the blood will increase if the kidneys are unable to eliminate creatinine from the blood. In most cases, calcium and phosphorus act as a buffer. By depositing phosphorus on the bones and other tissues, excessive phosphorus levels may cause low serum calcium as renal disease progresses. Because of their ability to cling to renal epithelial cells' surfaces, uric acid crystals may also cause an immediate inflammatory reaction. An increased risk of kidney stones and a decreased GFR are other possible side effects. Hyperparathyroidism and renal bone disease are often diagnosed by measuring the amount of serum ALP. A high ALP level in the blood of a CKD patient often implies rapid bone turnover.

BUN is a measure of the amount of urea nitrogen created in the body as a result of the breakdown of protein. Urine is the means through which it is expelled from the body. A rise in BUN may be caused by illness, injury for the kidneys. A buildup of potassium is a common side effect of renal failure since the kidneys are unable to eliminate it from the body. Hyperkalemia, or high potassium levels in the blood, is a complication of severe stages of chronic renal disease⁹. Patients with renal illness have abnormal serum enzymes (Alanine amino Transferases). The lack of renal excretion and the frequent occurrence of several comorbid diseases are two factors that contribute to this. Kidney stones and crystals may develop when an excessive quantity of oxalate in the urine combines with calcium. Renal failure leads for a recurring kidney stones and crystals.

Table 2 : Effect on oral administration of NNE, ALE, LAE and PHF on Ethylene Glycol (0.75% v/v)

Groups	Treatment	Descriptive Tests			
		BUN (mg/dL)	Potassium (mEq/L)	Alanine Amino Transferases (U/L)	Oxalate (mg/dL)
Normal	Saline	19.46±0.42	3.54±0.09	16.46±0.24	159.9±0.48
Control	Ethylene glycol (0.75%v/v)	29.75±0.47	5.53±0.16	49.29±0.3	796.4±3.08
Standard control	Cystone (750 mg/kg)	21.45±1.41	3.45±1.04	20.76±1.47	386.9±0.58
Test Group I	NNE (100mg/kg, p.o.)	25.94±0.27	4.77±0.06	34.07±0.52	374.5±0.42

Test Group II	<i>NNE</i> (200mg/kg, p.o.)	18.76±0.3	3.76±0.075	24.91±0.35	319.2±0.3
Test Group III	<i>ALE</i> (250mg/kg, p.o.)	27.58±0.41	4.9±0.028	37.41±0.24	485.5±0.42
Test Group IV	<i>ALE</i> (500mg/kg, p.o.)	23.45±0.41	4.57±0.048	29.53±0.4	327.6±0.41
Test Group V	<i>LAE</i> (250mg/kg)	28.24±0.88	3.63±0.09	33.03±0.46	361.8±5.04
Group VI	<i>LAE</i> (500mg/kg)	18.94±0.38	3.165±0.01	23.10±0.48	317.4±1.68
Group VII	<i>PHF</i> (250mg/kg)	24.85±0.69	4.03±0.024	37.10±0.58	492.3±2.10
Group VIII	<i>PHF</i> (500mg/kg)	25.29±1.43	3.02±0.04	21.59±1.73	397.3±1.84

NNE, *ALE*, *LAE* and *PHF*: Methanolic *Nelumbo nucifera*, *Anogeissus latifolia*, *Leucas aspera* and *PHF* extracts at which *P<0.05 & 0.01 is taken for Tukeys test of significance.

Table 3 : Methanolic Extracts of *NNE*, *ALE*, *LAE*, *PHF* on (0.75%v/v) EthyleneGlycol Induction on Urolithic Rats.

Group	Treatment	Descriptive Tests		
		Lactate dehydrogenase (nmole/min/mL)	Glutathione Stimulating Hormone (GSH) (nmol/mg of protein)	Lipid Peroxidation (LPO) (nmoles of MDA/g protein)
Normal control	Saline	353.6±0.58	4.58±0.008	1.52±0.006
Toxic Control	Ethyleneglycol (0.75%v/v)	749±0.36	1.46±0.002	2.95±0.004
Standard control	Cystone (750 mg/kg)	457.1±0.26	4.72±0.01	1.34±0.006
Test Group I	<i>NNE</i> (100mg/kg)	577.9±1.49	4.99±0.05	1.47±1.007
Test Group II	<i>NNE</i> (200mg/kg,b.wt,p.o.)	473.5±0.55	4.65±0.01	1.23±0.006

Test Group III	<i>ALE</i> (250mg/kg,b.wt.p.o.)	598.1±0.54	3.59±0.004	1.93±0.01
Test Group IV	<i>ALE</i> (500mg/kg,b.wt.p.o.)	486.9±0.33	4.37±0.01	1.62±0.007
Test Group V	<i>LAE</i> (250mg/kg,b.wt.p.o.)	567.5±1.43	3.73±0.077	1.587±0.004
Test Group VI	<i>LAE</i> (500mg/kg,b.wt.p.o.)	459.3±2.4	4.593±0.068	1.282±0.013
Test Group VII	<i>PHF</i> (250mg/kg,b.wt.p.o.)	566.8±0.87	3.69±0.023	2.03±0.01
Test Group VIII	<i>PHF</i> (500mg/kg,b.wt.p.o.)	436.3±1.74	4.49±0.12	1.373±0.02

NNE, *ALE*, *LAE* and *PHF*: Methanolic *Nelumbo nucifera*, *Anogeissus latifolia*, *Leucas aspera* and *PHF* extracts at which * $P < 0.05$ & 0.01 is taken for Tukeys test of significance.

Figure A: Eth.G. induction in rats after 28 days oral administration of *NNE*, *ALE*, *LAE* and *PHF*.

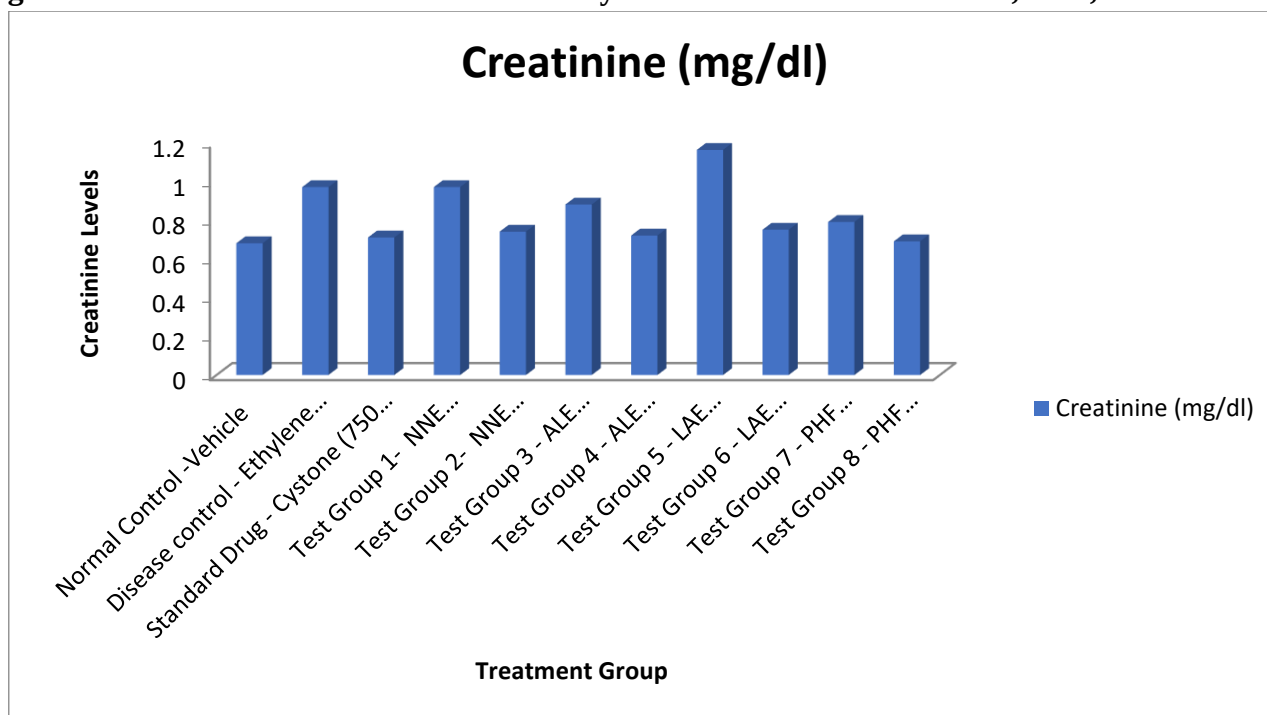


Figure B: Rats with Ethylene Glycol-induced Urolithiasis were treated with oral administration of *NNE*, *ALE*, *LAE* and *PHF* for 28 days before calcium levels were measured.

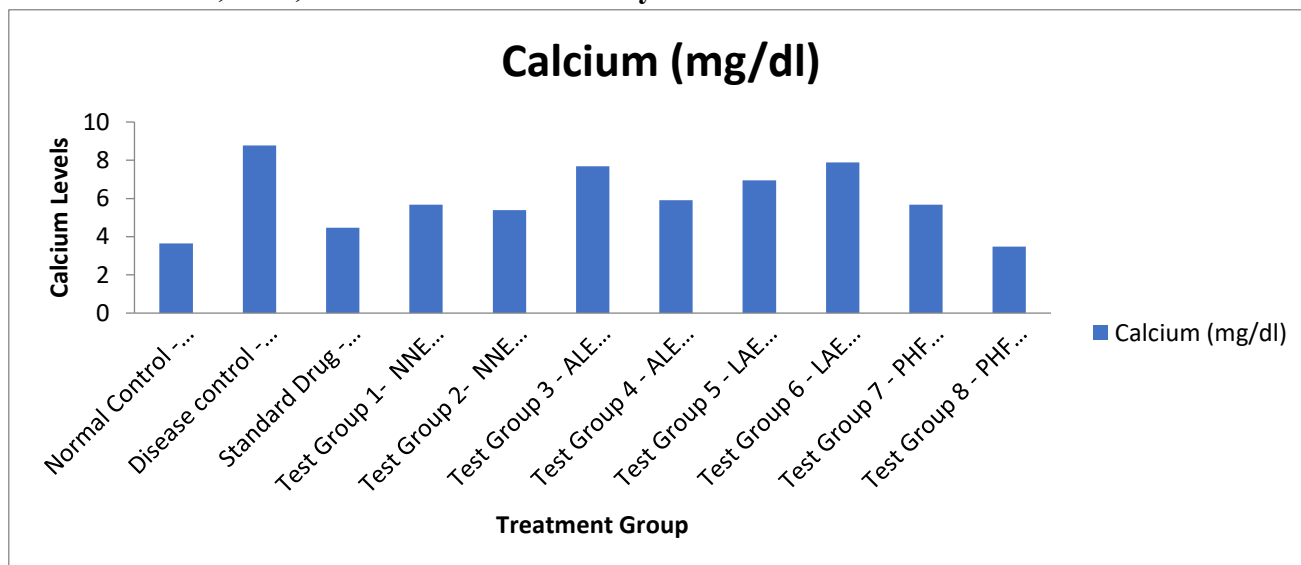


Figure C: Methanolic Extracts of *NNE*, *ALE*, *LAE* and *PHF* on Eth.G. with Phosphorus Analysis after 28 days of treatment.

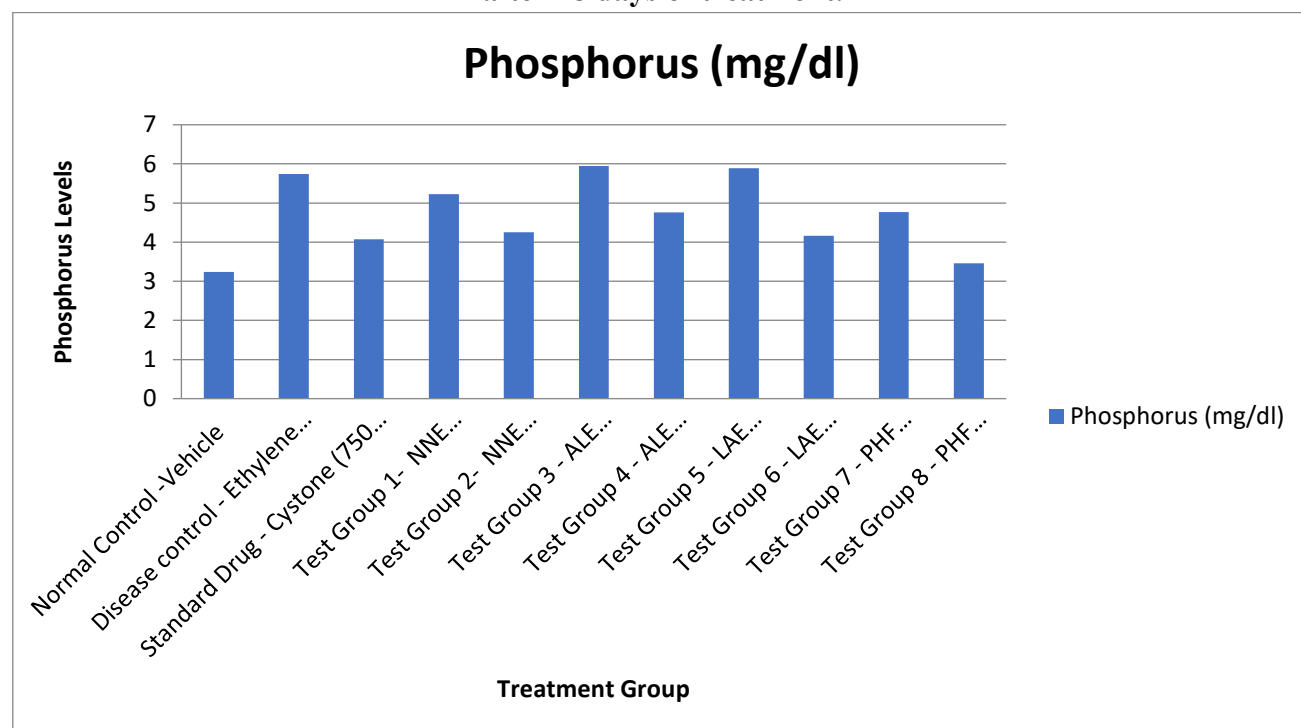


Figure D: Methanolic Extracts of *NNE*, *ALE*, *LAE* and *PHF* on Eth.G. with Uric Acid after 28 days of treatment.

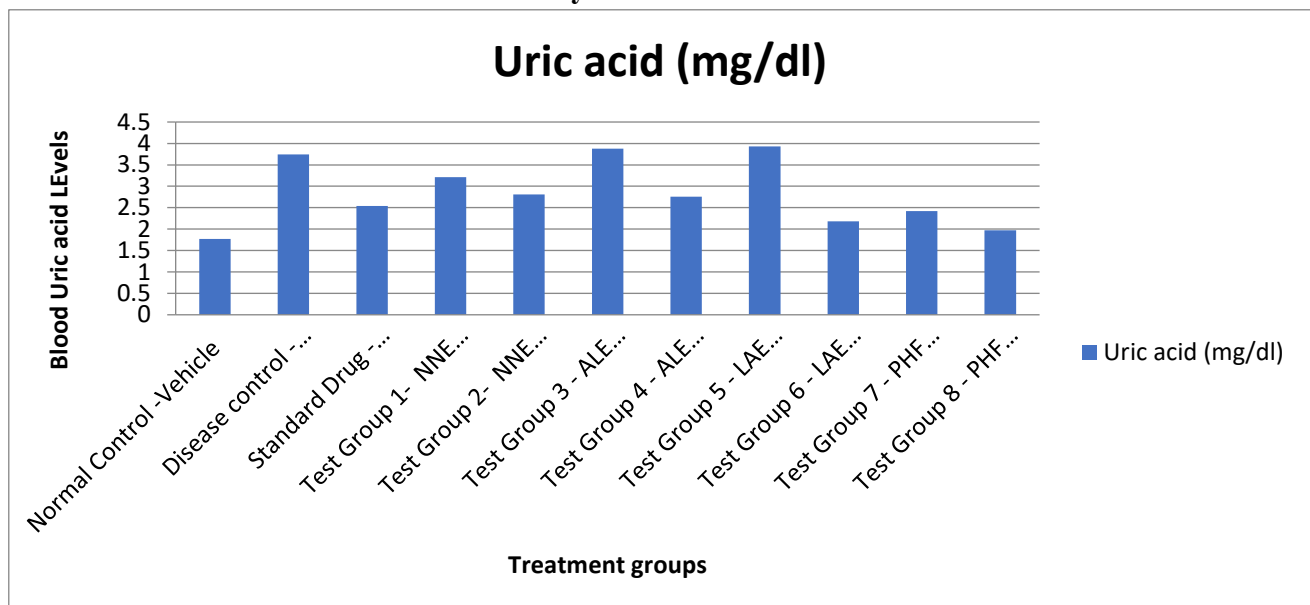
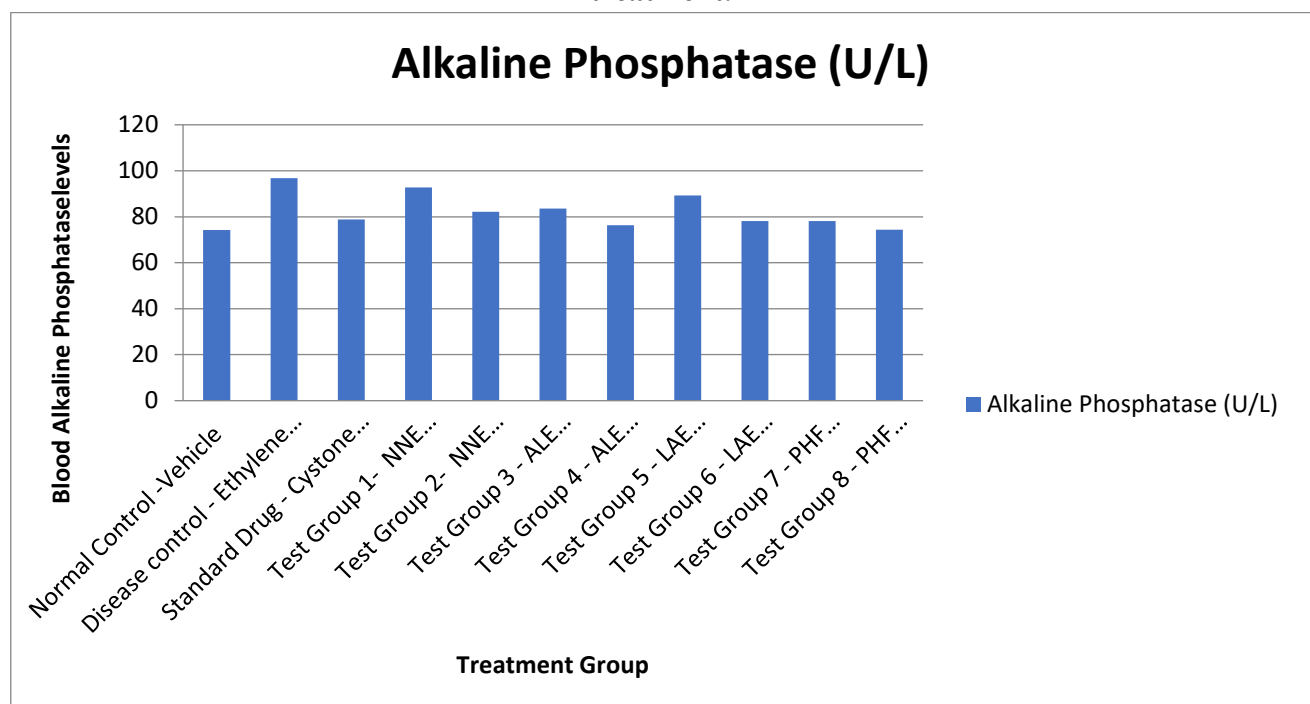


Figure E: Methanolic Extracts of *NNE*, *ALE*, *LAE* and *PHF* on Eth.G. with ALP after 28 days of treatment.



LDH plays an important role in the energy production in the body. A rise in LDH levels is diagnostic of tissue damage. It is released into the circulation as well as any other body fluids that may be present. Glutathione (GSH) is critical to the kidneys' ability to function normally. Antioxidants that protect cells from ROS and other poisons are the most important hydrophilic antioxidants. Because aldehydes are crucial signaling intermediates in altering cellular physiological function by influencing numerous transcription factors, they play a significant role in illness. Lipoprotein peroxides that are too high may suggest a deficiency in CoQ₁₀. Oxidative stress may lead to tissue damage and an increased risk of illness.

if it persists for an extended period of time.

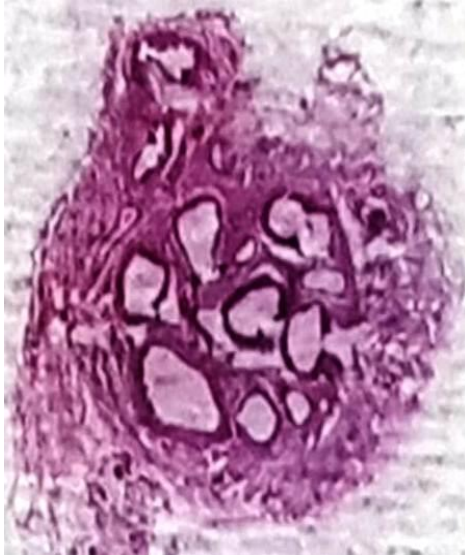
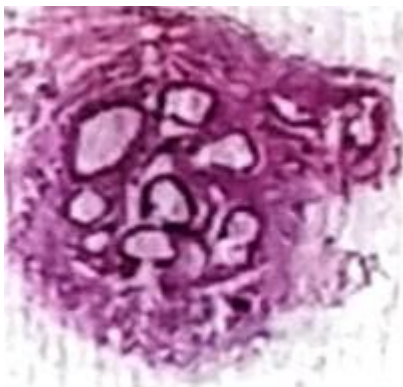
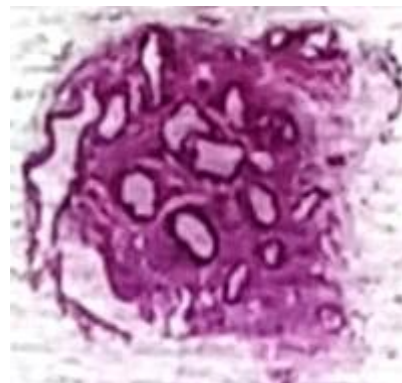
6.2 KIDNEY HOMOGENATE ANALYSIS

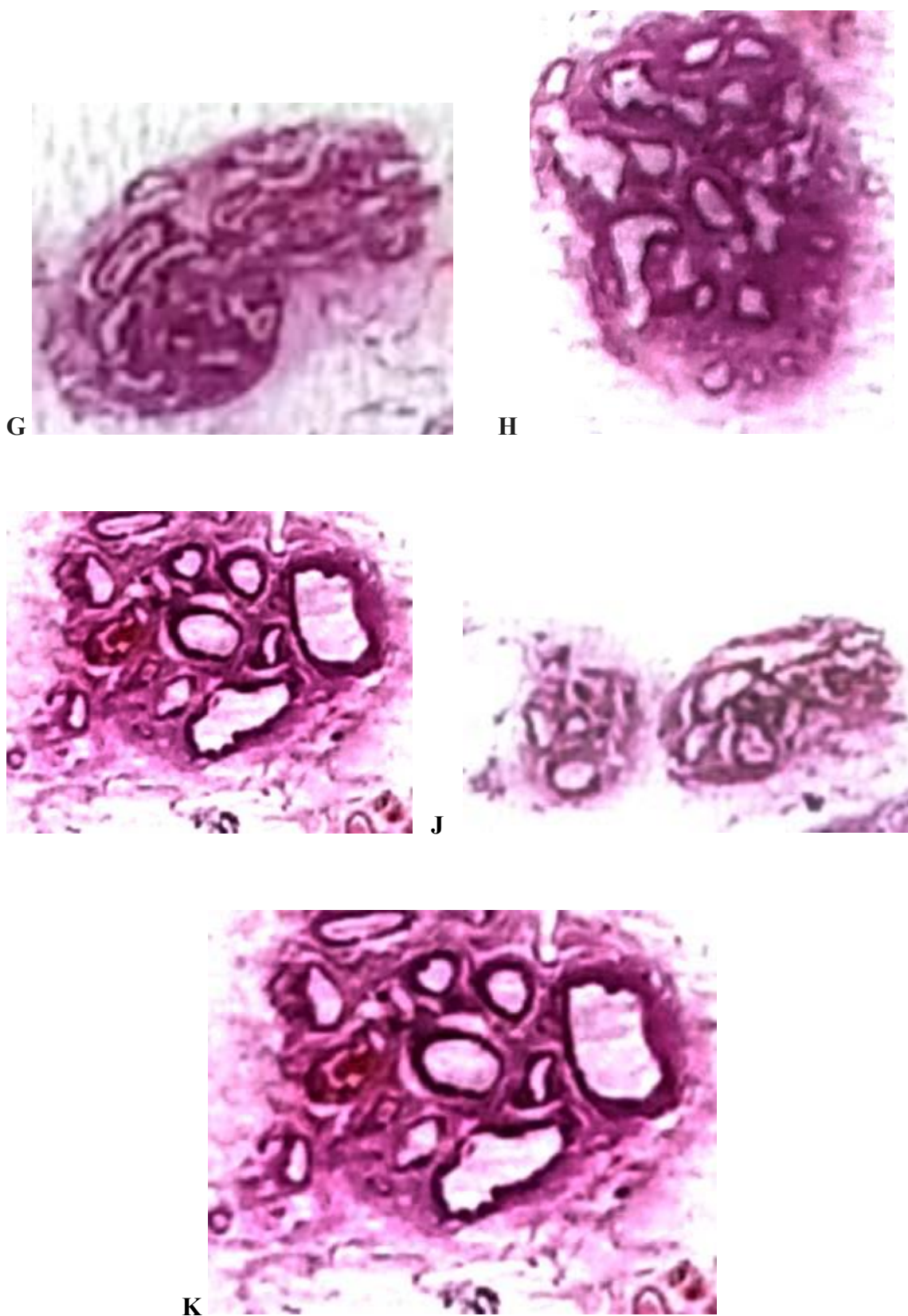
Physiological saline that had been stored in the freezer was used to clean out the rats' kidneys after they died on day 28 from large doses of pentobarbital and remove any superfluous tissue. Two kidneys had been cleaned and dried in an air oven at 80 degrees Celsius; their dry weight was also noted. Both kidneys, one on each side, were seen here. The right kidney was preserved in formalin at a concentration of 10 percent for a total of two days. In order to get rid of the formalin, the kidney was submerged in running water for about 12 hours, and then it was dehydrated with progressively stronger concentrations of isopropyl alcohol for a total of 12 hours (70, 80, and 90 percent). The last phase is to dehydrate yourself with alcohol that is one hundred percent strength for a total of twelve hours. This should be done about three times. The process of dehydration was carried out so that any remaining traces of water may be removed. After removing the last of the alcohol using chloroform, the technician employed paraffin infiltration to get rid of the chloroform residue. Two applications of chloroform, each lasting between 15 and 20 minutes, were utilised to thoroughly clean the region. After the paraffin infiltration, the slices of tissue were examined using a staining method that consisted of hematoxylin and eosin. The solid paraffin was melted before being poured into blocks in the form of an L. Almost immediately after that, the kidney chunks were dropped into the molten paraffin, and they were left there until the paraffin cooled. Thin portions of the tissue were cut out and put on a microscope slide while using a micro slide that had been coated with egg albumin. At a temperature of 600 degrees Celsius, the components were cooked for one hour. The coagulation of paraffin and egg albumin is the process that is used to adhere tissue to a slide. This is because eosin is an acid stain, which means that it gives the cytoplasm a pink colour. All of the cell's acidic components appear blue when stained with haematoxylin, a basic stain. The kidney and calcium oxalate deposits were examined using polarised light and a light microscope.

6.3 HISTOPATHOLOGICAL STUDIES OF KIDNEYS

Cytoplasm of columnar epithelial cell stains in the coloration of pink in the tubules of Bowman's capsule at which glomerulus seems to be normal. Bowman's capsule surrounds glomerular degeneration with loss of capillaries. Degeneration and loss of tubular architecture are clearly visible in the tubules, as is the buildup in the tubules' centres. Bowman's capsule surrounds a glomerulus that has suffered a loss of capillaries. A minor Tubular degeneration is evidenced by build up in the tubules' centres when the tubules recover from toxicity. Bowman's capsule surrounded by a tuft of capillaries shows that the glomerulus has recovered its usual structure. The vast majority of tubules have returned to their usual structure and function. A few tubules, however, seemed to have modest degeneration, as seen by the buildup of fluid in the core. Bowman's capsule surrounded by a tuft of capillaries shows that the glomerulus has recovered its usual structure¹⁰. The tubules had a normal structure and were recovering normally. Bowman's capsule surrounded by a tuft of capillaries shows that the glomerulus has recovered its usual structure. The vast majority of tubules have returned to their usual structure and function. Few tubules, on the other hand, had modest degeneration, as shown by accumulation in the tubules' centres. Bowman's capsule surrounded by a tuft of capillaries shows that the glomerulus has recovered its usual structure. The tubules had a normal morphology and were recovering at a modest pace. Glomerulus can be with a tough capillaries surrounded by bowman's capsule. Tubules were

encircled with an unremarkable Blood Vessels. Interstitium will be unremarkable followed by a tough capillaries inside the glomerulus surrounded by Bowman's capsule.

**A****B****C****D****E****F**



7. DISCUSSION

6.87 percent Saponin of Ethanolic Extract of *Nelumbo nucifera*, 0.5 185gm of Saponin was recovered from 5gms of crude extract. Saponin was discovered to have a percentage of 10.37. *Linn* Seed of

Anogeissus latifolia, *Leucas aspera* was 16.14 percent. By quantitative determination, 0.7549 of Saponin was recovered from 5gm of crude extract. Saponin content was found to be 15.09 percent. It was established that saponins derived from the rhizomes of *Anogeissus latifolia*, *Leucas aspera*, and *Nelumbo nucifera*. It had no acute oral toxicity when administered to Wistar rats. Rhizome of *Anogeissus latifolia* Linn Seeds are suggested therapeutic dosages by OECD 425. Kidney failure is indicated by an elevated creatinine level. Hypomagnesemia, hypercalciuria, and hyper-phosphaturia were the disorders caused by the induction of ethylene glycol. The creatinine level in the blood will increase if the kidneys are unable to eliminate creatinine from the blood. In most cases, calcium and phosphorus act as a buffer. By depositing phosphorus on the bones and other tissues, excessive phosphorus levels may cause low serum calcium as renal disease progresses. Uric acid crystals may cling to the surface of the renal epithelial cells, causing an immediate inflammatory reaction. In addition, kidney stones and a decreased glomerular filtration rate are also possible side effects (GFR). Serum ALP concentrations are often used to identify hyperparathyroidism and renal bone disease. When a CKD patient's blood ALP levels are elevated, it frequently means that their bones are resorbing quickly. The amount of urea nitrogen that is produced in the body as a consequence of the breakdown of protein is the quantity that is measured by BUN. It is (ultrahigh dosage) (high dose). Eliminated from the body through the urinary system, namely the urine. An increase in BUN may be the consequence of a decline in renal function brought on by disease or damage to the kidneys. This may be the case in certain cases. For patients with renal failure, a buildup of potassium is a common complication that occurs when the kidneys cannot eliminate the excess. Hyperkalemia, or high potassium levels in the blood, is a possible consequence of severe renal disease.

8. CONCLUSION

Serum enzyme levels are aberrant in patients suffering from renal disease (Alanine amino Transferases). Two reasons that contribute to this are the absence of renal excretion and the frequent incidence of numerous disorders. Kidney stones and crystals may develop when an excessive quantity of oxalate in the urine combines with calcium. Renal failure occurs if kidney stones and crystals continue to accumulate regularly. When it comes to producing energy in the body, LDH plays an important function. Tissue injury is indicated by an increase in LDH. As an example: Many tissues, including the blood and heart as well as the kidneys and brain, might be affected. As a result of any damage to these tissues, LDH and other body fluids are discharged into the bloodstream.

Glutathione Stimulating Hormone (GSH) is critical to the kidneys to function normally. Oxidative stress may lead to tissue damage and an increased risk of illness if it persists for an extended period of time. Tests were performed that were able to control the development of calcium oxalate crystals by aggregating. It was possible to see both the crystallization of calcium oxalate and its prevention using microscopy. Bowman's capsule tends to have a loss of capillaries along with the degeneration of glomerulus, according to histological studies. tubular degeneration, tubular morphological cell and tissue damage loss occurs and minimised by a conventional medication by cystone, respectively, are seen.

Both the low doses of the both the drugs demonstrate normal architecture of glomerulus and has normal recovery of cells and tissues in the tubules. Both high doses of extracts shows recovery and normal architecture of glomerulus the tubules were showing normal architecture and recovery. Methanolic forms of *Nelumbo nucifera*, *Anogeissus latifolia*, and *Leucas aspera* tends to have an anti-urolithic action can be

exerted by inhibiting the stone formation followed by damage to the internal surfaces of the renal tubules of Bowman's capsule and glomerulus in the kidneys. Thus, it can be enhanced that the recovery and growth of the cells and tissues can be seen with the replacement of degenerative and necrotic areas in the renal system.

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Nil.

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9. REFERENCES

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