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Research Article

**EVALUATION OF *IN VITRO* ANTI UROLITHIATIC ACTIVITY
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J. Himabindhu***Department of Pharmacognosy, Vishnu Institute of Pharmaceutical Education and Research,
Narsapur, Medak, Telangana.**Abstract:**

The present study was explores that evaluation of in vitro antiurolithiatic activity of Senna occidentalis. It was observed that the highest calcium oxalate crystals dissolution was observed in the ethanolic extract of Senna occidentalis. It was found that ethanolic extract of Senna occidentalis has more efficient to dissolve calciumoxalate. In this study Neeri was used as standard drug.

Key words: *Senna Occidentalis, urolithiasis and Ethanolic extract*

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INTRODUCTION:

Kidney stones are a common cause of blood in the urine and pain in the abdomen, flank, or groin. Kidney stones occur in 1 in 20 people at some time in their life. The development of the stones is related to decreased urine volume or increased excretion of stone-forming components such as calcium, oxalate, urate, cystine, xanthine, and phosphate. The stones form in the urine collecting area (the pelvis) of the kidney and may range in size from tiny to staghorn stones the size of the renal pelvis itself. The process of stone formation, urolithiasis, is also called nephrolithiasis.

The worldwide incidence of urolithiasis is quite high, and more than 80% of urinary calculi are calcium oxalate stones alone or calcium oxalate mixed with calcium phosphate [1]. However, the presence of certain molecules raise the level of supersaturation of salts needed to initiate crystal nucleation or reduce the rate of crystal growth or aggregation and prevents stone formation [2]. Calcium oxalate stones represent up to 80% of analyzed stones [3]. Calcium phosphate account for 15-25%, while 10-15% is mixed stones. The others are struvite 15-30%, cystine 6-10%, and uric acid stones 2-10% [4]. Calcium oxalate stones are of primary two types, calcium oxalate monohydrate (whewellite) and calcium oxalate dihydrate (weddellite). The occurrence frequency of whewellite is 78% while that of weddellite is 43% [5]. Though technological advancements have made dramatic improvement in the removal of urinary stones still some of the drawbacks of these methods exists which includes their being too costly for a common man and recurrence of stone formation along with a number of other side effects [6]. Many medications and remedies have been used during the past many years to treat urinary stones. Endoscopic stone removal and extracorporeal shock wave lithotripsy have revolutionized the treatment of nephrolithiasis, but do not avoid the possibility of new stone formation [7]. Various therapies including thiazide diuretics and alkali citrate are being used in an attempt to prevent the recurrence of hypercalciuria and hyperoxaluria induced calculi, but scientific evidence for their efficacy is less convincing [8].

Medicinal plants have played a significant role in various ancient traditional systems of medication. Even today, plants provide a cheap source of drugs for the majority of the world's population. Several pharmacological investigations on the medicinal plants used in traditional antiurolithiatic therapy have revealed their therapeutic potential in their *in-vitro* or *in-vivo* models [9].

Senna occidentalis is an erect foetid annual herb. It belongs to the family Fabaceae and commonly called as coffee Senna and in English; it is called as septic weed. It grows up to 60 to 150 cm in height and it is found throughout India up to an altitude of 1500 cm. *Senna occidentalis* has many traditional uses to treat typhoid, malaria, dog bites. It has pharmacological activities like antihelmintic [10], antifungal, antimutagenic, antipyretic and antifeedant. In this article we present an Anti Urolithiatic activity of Ethanol extract of *Senna occidentalis* by using Titrimetry method.

MATERIALS AND METHODS:**Plant Materials**

The leaves of *Senna occidentalis* were collected from Khagazmaddur (Vil), Narsapur (Mdl), Medak (Dist) of Telangana in the month of August 2017. The plant was authenticated by D. Venkateshwar Rao, Deputy Director, Telangana Forest Academy, Dullapally, Hyderabad, Rangareddy District. The leaves were washed with tap water and dried under shade.

Preparation of Plant Extract

The leaves of plant were dried under shade and crushed in pulveriser and powdered. These powdered plant material was extracted with Ethanol in a Soxhlet apparatus for 72 hours. After complete the extraction, the extracts were cooled at room temperature and filtered and evaporated to dryness using rotary evaporator.

Chemicals Used

Neeri, Sodium oxalate, Tris buffer, calcium chloride, Potassium permanganate (KMnO₄), Sulphuric acid (H₂SO₄).

Investigation of *In Vitro* Antiurolithiatic Activity Test by Titrimetry

The experimental kidney stones of calcium oxalate (CaOx) were prepared in the laboratory by taking equimolar solution of calcium chloride dehydrate in distilled water and sodium oxalate in 10 ml of 2N H₂SO₄. Both were allowed to react in sufficient quantity of distilled water in a beaker, the resulting precipitate was calcium oxalate. The precipitate was freed from traces of sulphuric acid by ammonia solution, washed with distilled water and dried at 60°C. The dissolution percentage of calcium oxalate was evaluated by taking exactly 1 mg of calcium oxalate and 10 mg of the extract, packed together in semipermeable membrane of egg as shown in the model designed given below. This was allowed to suspend in a conical flask containing 100 ml of 0.1M Tris buffer. First group served as blank

containing only 1 mg of calcium oxalate. The second group served as positive control containing 1 mg of calcium oxalate and along with the 10 mg standard drugs, i.e. Neeri. The 3rd group along with 1 mg of calcium oxalate contain Ethanolic extracts. The conical flasks of all groups were kept in an incubator preheated to 37 °C for 2 h. Remove the contents of semipermeable membranes from each group into separate test tubes, add 2 ml of 1N sulphuric acid to each test tube and titrated with 0.9494 N KMnO₄ till a light pink colour end point obtained. The amount of remaining undissolved calcium oxalate is subtracted from the total quantity used in the experiment in the beginning to know the total quantity of dissolved calcium oxalate by various solvent extracts¹¹.

RESULTS AND DISCUSSION:

In the present study, Titrimetry method was used to assess the antiurolithiatic activity of Ethanolic extract of *Senna occidentalis*. The dissolution percentage, i.e. 51% of calcium oxalate (CaOx) dissolution was observed in Ethanolic extract. From this study, it was observed that Ethanolic extract of *Senna occidentalis* leaves showed antiurolithiatic activity. This study has given primary evidence for *Senna occidentalis* the plant which possess lithotriptic property. This in vitro study has given lead data and shown that Ethanolic extract of *Senna occidentalis* is quite promising for further studies in this regard.

Table 1: Shows % dissolution of calcium oxalate (CaOx) by in vitro anti urolithiatic activity of *Senna Occidentalis* leaves extracts.

Sl.No.	% of dissolution of calcium oxalate	
	Groups	<i>Senna Occidentalis</i>
1.	Blank	0
2.	Positive control	81
3.	Ethanol extract	51



Figure 1(a): Decalcification of egg shell in 10% Acetic acid overnight.



Figure 1(b): Decalcified Eggs

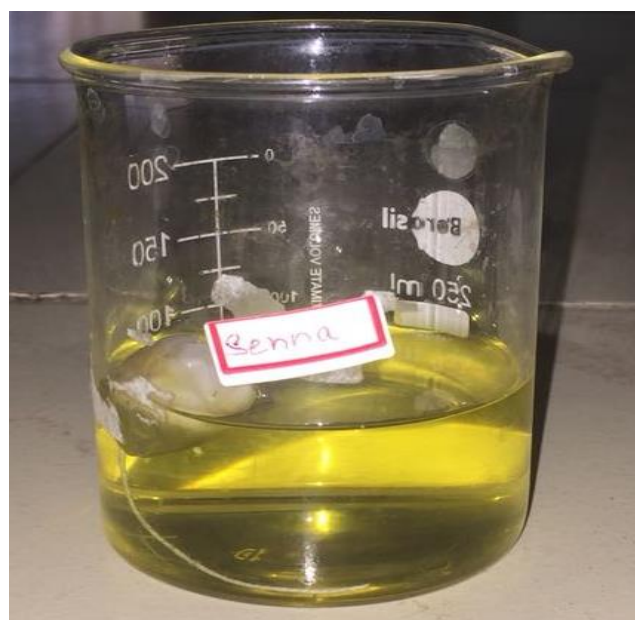


Figure 1(c): Egg membrane along with the contents suspended into the 0.1 M Tris buffer.

Figure 1: In vitro experimental model setup to evaluate antiurolithiatic activity

CONCLUSION:

In the present work, the dissolution of calcium oxalate crystals by Ethanolic extract of *senna occidentalis* was studied by using the standard drug, cystone. The work was performed by using in vitro antiurolithiatic model for calculating percentage dissolution of kidney stone. This study has given primary evidence for *Senna occidentalis* as the plant which possess antiurolithiatic property.

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