INHIBITION OF CALCIUM OXALATE CRYSTALLIZATION IN-VITRO BY CLITORIA TERNATEA ROOT
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ABSTRACT
Clitoria ternatea, commonly known as Aparajita (family: Fabaceae). The inhibition of in-vitro calcium oxalate crystal (a common major component of most urinary stones) formation by various extract of Clitoria ternatea was investigates by titrimetric method. The inhibitor potency of alcoholic extract of Clitoria ternatea was found to be comparable to that of Cystone (a proprietary drug for dissolving kidney stones). Thus alcoholic extract could be further analyzed in vivo and further characterized of its active compound could lead to the discovery of a new candidate drug for the patient with Urolithiasis.

INTRODUCTION
Lithiasis is the formation of calculi or stone which is a concretion of material mainly salt in any part of body. Oxalate is a metabolic end product and a major constituent of the majority of renal stones. Extract of Clitoria ternatea root has been found to be having Anti-lithiasis activity.

MATERIAL AND METHOD
The part of Clitoria ternatea was collected from Jodhpur, Rajasthan. During the month of January and February and plant was identified & confirmed with the herbarium of BSI. Plant authentication no. is 12012/20-10-2011/TECH (Pl.id)/600.

Extraction-Fresh plant after collection was shade and dried and then the powdered leaves were extracted with alcohol by soxhlet apparatus while aqueous extract was obtained by seven day maceration.

Preparation of experimental kidney stones (calcium oxalate) by homogenous precipitation-Equimolar solution of calcium chloride dehydrates (AR) in distilled and sodium oxalate (AR) in 10 ml. of 2N H₂SO₄ were allowed to react in sufficient quantity of distilled water in a beaker. The resulted precipitate was calcium oxalate.

Preparation of semi permeable membrane from farm eggs-The semi permeable membrane from eggs lies in between outer calcified shell and the inner content like albumin and yolk. Shell was removed chemically by placing the egg in 2M HCl for an overnight, which caused decalcification. Further washed with distilled water and carefully sharp pointer a hole is made on the top and the contents squeezed out completely from the decalcified egg washed thoroughly with distilled water and placed it in ammonia solution, in the moistened condition for a while and then rinsed it with distilled water, store in refrigerator at pH of 7-7.4.

Estimation of calcium oxalate by titrimetric analysis-Weighed exactly 1mg of the calcium oxalate and 10 mg of the extract and packed it together in semi permeable membrane by suturing. This was allowed to suspend in a conical flask containing 100ml of 0.1M of TRIS buffer. One group served as negative control (contained only 1mg of calcium oxalate). Placed the conical flask of all groups in incubator, pre heated to 37°C for 2 hours. For about 7-8 hours removed the content of semi
permeable membrane from each group into a test tube. Added 2 ml of 1N H₂SO₄ and titrated with 0.9494N KMNO₄ till a light pink colour end point is obtained. 1 ml of 0.9494N of KMNO₄ equivalent to 0.1898 mg of calcium. The amount of undissolved calcium oxalate is then subtracted from the total quantity used in the experiment in the beginning. To know how much quantity of calcium oxalate actually the test substance could dissolved.

RESULT AND DISCUSSION
Alcoholic extract of leaves of Clitoria ternatea showed higher calcium oxalate crystallization inhibition (72.99±1.2%) in vitro while cystone, a prescribed medicine for renal calculi showed highest inhibition (90.55±1.27%) in terms of formation of calcium oxalate precipitation.

CONCLUSION
Thus present study demonstrated that alcoholic extract of leaves of Clitoria ternatea showed comparable activity to that of cystone in terms of inhibiting the formation of calcium oxalate precipitate. The Alcoholic extract of root of Clitoria ternatea could be further analysed in vivo and further characterization of its active compound could lead to a new candidate drug for patients with Urolithiasis.

REFERENCES