Evaluation of Anti-Urolithiasis Activity in Coffea Arabica.Linn using Struvite Crystal Growth Inhibition Assay

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ABSTRACT
Struvite accounts for 15 – 20% of all renal calculi. These stones can grow rapidly forming ‘straghorn calculi’, which is a more painful urological disorder threatening human life particularly the women. The present work was aimed to grow struvite crystals in vitro using single diffusion gel growth technique and to understand the effect of Coffea arabica on its growth. Test drug was prepared at two different concentrations of 0.5 and 1% dispersed in 1.0M magnesium acetate solution were gently poured on the set gel in the test tubes to enumerate the growth inhibition of struvite crystals. The result showed that the test drug Coffea arabica has anti –urolithiasis property in the tested medium.

Keywords: struvite, Coffea Arabica, crystallization, straghorn stone

INTRODUCTION
Struvite crystallization is related to urinary tract infection by microorganism producing urease. They are mainly the microorganism from the species of proteus. The techniques for removal of calculi such as, endoscopic stone removal lithotripsy and extra corporal shock wave lithotripsy cause traumatic effect of shock wave. Which leads to decrease in renal function. Therefore antilithic drugs from natural sources have assumed greater importance as herbal alternative which are cost effective with low side effects.

These Coffea arabica were claimed to cause no bacterial resistance and other adverse effects.

However only a few studies have been conducted on the effect of the herb on the crystal growth of struvite as of the main components of renal calculi. Coffea arabica was known to possess several pharmacological effects mainly as, antioxidant, anti-inflammatory, antimicrobial, diuretic, lithotriptic etc. Keeping in view the importance of Coffea arabica, the present work was aimed to study its influence on the growth of struvite crystals in vitro.

MATERIAL AND METHODS:

COLLECTION OF RAW DRUG:
Raw drug were collected from kodaikanal, these drug were authenticated by Medicinal Botany department, Arumbakkam, Chennai -106. (GSMC/MB- Voucher specimen no.39/2017). Project Id: NRS/AS/0061/06/2017.

BOTANICAL CLASSIFICATION:

Kingdom: Plantae
Clade: Angiosperms
Order: Gentianales
Family: Rubiaceae
Genus: Coffea
Species: Arabica.
VERNACULAR NAMES OF COFFEA ARABICA:[6]

- Tamil: Kappikotti
- English: seeds of Coffee, Arabian coffee
- Telugu: Kapi-vittulu
- Malayalam: Kappik-kuru
- Kanata: Kapi-bija
- Sanskrit: Meecha-phala.1
- Arab: Bun
- Hindi: Bun.

METHOD OF PREPARATION:

Seeds of Coffea arabica are roasted then make it fine powered.

ANTI-UROLITHIATIC ACTIVITY:[7]

Objective

The single diffusion gel growth technique was adopted to evaluate anti-urolithiatic potential of the study drug Coffea arabica

Test Drug concentration

Test drug was prepared at two different concentrations of 0.5 and 1% dispersed in 1.0 M magnesium acetate solution

Methodology

An aqueous solution of 0.5M Ammonium dihydrogen phosphate was admixed with the sodium metasilicate solution of specific gravity 1.05 in appropriate amount using magnetic stirrer so that the pH value 7.0. pH of the reaction was ensured by using pH probe meter. The gel solution of 10 mL was transferred into the test tubes of 140 mm length and 25 mm diameter. After the gelation took place, 5 mL of supernatant solutions of 0.5 and 1% conc of test drug in 1.0 M magnesium acetate were gently poured on the set gels in test tubes to enumerate the growth inhibition of Struvite crystals. About 5 ml of 1.0 M magnesium acetate without test drug were added as supernatant to control tubes which serves as crystal control group. All the procedures was done in the aseptic medium in laminar flow hood to avoid microbial contaminations. All test tubes and other glassware were autoclaved at 120°C for 15 min. After pouring supernatant solution, the test tubes were capped with airtight stopples. The experiment was conducted at the room temperature. Study on growth of crystal were carried out for five consecutive days.

Growth Pattern of crystal in control and drug added medium
Size variation of Struvite crystals

A - Size variation of Struvite crystals in Control Gel medium
B - Size variation of Struvite crystals in Gel medium with 0.5 % of Coffea arabica
C - Size variation of Struvite crystals in Gel medium with 1 % of Coffea Arabica

Microscopic view of Struvite crystals size after fragmentation.

RESULTS AND DISCUSSION:
The present study investigated the anti-uroolithiasis activity of Coffea Arabica using struvite crystal growth inhibition assay. This results were tabulated.
Report on Average Length of the Crystal in different medium

<table>
<thead>
<tr>
<th>S.No</th>
<th>Medium</th>
<th>Average Length of the Crystals in cm</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control Gel medium</td>
<td>Mean: 1.44</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Deviation: 0.3362</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Error: 0.1503</td>
</tr>
<tr>
<td>2</td>
<td>Gel medium + 0.5 % CA</td>
<td>Mean: 0.72</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Deviation: 0.1304</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Error: 0.05831</td>
</tr>
<tr>
<td>3</td>
<td>Gel medium +1% CA</td>
<td>Mean: 1.04</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Deviation: 0.2608</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Std. Error: 0.1166</td>
</tr>
</tbody>
</table>

Observation

Control Medium
Average size of the crystal was higher in the control medium with the Avg length of 1.44 cm.

Gel medium + 0.5 % CA
Average size of the crystal was significantly decreased in medium contains 0.5% of test drug Coffea arabica with the Avg length of 0.72 cm.

Gel medium +1% CA
Average size of the crystal was much reduced in medium contains 1 % of test drug CP with the Coffea arabica length of 1.04 cm.

From the result of the study it was concluded that the test drug Coffea arabica has Promising anti-urolithiasis property in the tested medium.

CONCLUSION:
The stone formation process occurring in the human body is quite complex, and takes place in a dynamic environment; from the present study one can suggest that the seeds of Coffea Arabica Linn. Inhibit the growth of struvite crystals in vitro. This study may be may be used for formulating the strategy for prevention or cure.

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