

Antibacterial efficacy of Apamarga and Karanja kshara against *Staphylococcus aureus* and *Escherichia coli*

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Introduction and Objectives: Plant-derived antibacterials have played a crucial role in traditional medicine since ancient times. Currently, the world is facing emerging antibiotic resistance, thereby reducing the effectiveness of antibacterials. This study aimed to assess the in vitro antibacterial efficacy of Apamarga and Karanja kshara against *Staphylococcus aureus* and *Escherichia coli*.

Methods: Whole Apamarga (*Achyranthes aspera*; Karal heba; Pungai) and Karanja (*Pongamia pinnata*; Magul karanda; Naagarkaa mullu) plants were cut into pieces and after shade drying, were burnt in a copper vessel with 1/10th limestone. Ash was collected by sieving, dissolved in water (1:6), and filtered 21 times through cotton cloth. The filtrate (ksharajala) was heated until clear, red, sharp, and slimy. Subsequently, limestone (1/10th ash) was heated red hot at 800 °C for 1 hour. It was then macerated in a mortar and the powder was mixed with Ksharajala, heated until dry and ground to a fine powder. Kshara stock solutions (1000 mg/ml) were prepared by dissolving 10 g of Kshara powder in 10 ml of distilled water.

The pH of 10% aqueous solutions of Apamarga and Karanja kshara were checked using a pH meter. Broth microdilution method was used to determine the Minimum Inhibitory Concentrations (MIC) of Apamarga and Karanja kshara solutions against *Staphylococcus aureus* (ATCC25923) and *Escherichia coli* (ATCC25922). Two-fold dilution was carried out using Kshara stock solutions (1000 mg/ml) in sterile round-bottom 96-well plates using Luria-Bertani (LB) broth. Ciprofloxacin was used as the positive control for *E. coli* and clindamycin for *S. aureus*. Sterile distilled water was used as the negative control. Broth microdilution was performed in triplicate. MIC values were determined by streaking 1 µL from each dilution, positive control, and negative control well onto blood agar plates. Colony-forming units (CFU) were used to assess microbial growth, with zero CFU indicating positive result and one or more CFU indicating negative result.

Results: The pH values of 10% Apamarga Kshara and 10% Karanja Kshara aqueous solutions were 11.43 and 12.87 respectively. The MIC of Apamarga Kshara was 250 mg/ml against *S. aureus* and 62.5 mg/ml against *E. coli*, while the MIC of Karanja Kshara was 62.5 mg/ml against both organisms. Apamarga Kshara exhibited greater antibacterial activity against *E. coli* than *S. aureus*. Karanja Kshara showed lower MIC than Apamarga Kshara against *S. aureus*.

Conclusions: Both Apamarga and Karanja kshara have high pH values. Karanja kshara exhibited antibacterial activity against both *S. aureus* and *E. coli* while Apamarga kshara exhibited significant antibacterial activity against *E. coli*.

Keywords: Plant-originated antibacterials, Antibiotic resistance, traditional medicine, Apamarga Kshara, Karanja Kshara

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