Isolation of antagonistic *Lactobacillus* from cow dung and its nutritional improvement as probiotic strain

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Different strain of *Lactobacilli* spp. were isolated from fresh cow dung and designated as A1 to A10. Selected *Lactobacillus* isolates were then inoculated with different nutritional sources like MRS broth, Packed milk, Sugarcane juice, Mousammi juice and MRS broth + Sugarcane juice and MRS + Mousammi juice. Their antagonistic effect was tested against three selected pathogenic bacteria namely *S. aureus, E. coli* and *Pseudomonas* spp. The result clearly showed that some isolates when inoculated with MRS broth + sugarcane juice inhibited the growth of pathogenic strains in varying degree. Isolate A-6 and A-10 were found to be more effective in reducing growth of all three pathogenic species from 10 to 30 mm. Other isolates (A1 and A2) from the same source, were effective against *E. coli* and *Pseudomonas* spp. with a zone of inhibition of 15 to 10, respectively but no zone of inhibition was seen against *S. aureus*.

Key words: Cow dung, Lactobacillus, Probiotic

Introduction

All free living animals including man do hosts for a society comprise an enormous number of individual. The normal microflora differs between animal species, body sites of individuals within the same species (Gilliland and Speck, 1977).

Human as well as animals normally are born sterile, but shortly after birth colonization begins and every location is filled up with the fittest microbes from the environment, thus creating a balanced ecological system. (Olotu Olanrewaju, 2007).

In recent years there is a great interest for viable microorganisms that promote or support a beneficial balance of autochthonous microbial population of gastrointestinal tract. The weight of intestinal flora is between 1 to 2 kg, housing 10¹⁴ bacteria which means that there are more living bacteria than there are cells in a normal body, which plays an important role in equilibrium of the intestinal ecosystem.

The word probiotic is a compound of Latin and Greek word, means "Favorable to life". Probiotics are live microorganisms, which are considered "Friendly germs" due to benefits to the colon and the immune system, when administrated in adequate amount confirm a health benefit on the host.

The Lactobacilli group of bacteria was the first probiotic. Milk, which contains Lactobacilli bacterium called as *Lactobacillus acidophilus* was also recommended in 1920s and 1930s for the treatment of

constipation and diarrhea. This species of probiotic adheres to the intestinal mucus and epithelial cells and boosts beneficial bacteria.

MATERIALS AND METHODS

The present study was designed to isolate antagonistic Lactobacilli from fresh cow dung, which is undigested residues of herbivorous matter and passed through the animal gut.

A total of 10 sample of fresh cow dung were collected from different locations of Dehradun, Uttarakhand, India.

Isolation of Lactobacilli was done by serial dilution method. Dilutions were made up to 10⁻⁵, 100 ml from each tube was spreaded on the MRS medium as this is selective media for Lactobacilli. These plates were then incubated at 37°C for 24 to 48 hrs.

Selected Lactobacilli isolates were inoculated in tubes containing sterilized milk. The tubes were observed for coagulation. Only those isolates which produce smooth curding from milk were selected for further study.

Indicator organisms, which include *S. aureus*, *E. coli*, and *Pseudomonas* spp. were used to assess the amount of antagonistic activity produced by isolates. Cell free filtrate preparation of isolates was done by centrifugation method and then was transferred to fresh sterilized tubes to check antagonistic activity against the pathogenic organisms by the agar well diffusion assay as described by Schillenger and Lucke (1998).

Selected Lactobacilli isolates were then inoculated