## Lab.2: Isolation of lactic acid bacteria (LAB) from some foods by pourplate method

Lactic acid bacteria (LAB) comprise an ecologically diverse group of microorganisms united by formation of lactic acid as the primary metabolite of sugar metabolism. These bacteria utilize sugars by either homo- or heterofermentative Pathways. LAB are described as nutritionally fastidious.

They are Gram positive usually non -motile, non-spore forming rods and cocci.

LAB were first isolated from milk and have since been found in such foods and fermented products as meat, milk products, vegetables, beverages and bakery products and have been detected in soil, water, manure and sewage, also LAB exist in human and animal as normal flora.

## Genera of LAB:

Lactobacillus, Leuconostoc, Pediococcus ,Streptococcus ,Aerococcus, Alloiococcus, Carnobacterium, Dolosigranulum, Enterococcus, Globicatella, Lactococcus, Oenococcus, Tetragenococcus, Vagococcus, and Weissella.

## Usages of LAB:

- 1. Used as a flavoring and texturizing agent.
- 2. Used as a preservative in food, they inhibit food spoilage and pathogenic bacteria and preserve the nutritive qualities of raw food material for an extended shelf life.
- 3. Added as starters in food.
- 4. Used as probiotics, they generally regarded as safe (GRAS).
- 5. LAB play an important role in processing animal feeds like silage.

## Procedure

- 1- Mash the sample of food.
- 2- Add 0.1 ml or 0.1 gram of the mash sample into the 10 ml of MRS broth, mix well and incubate at 37°C for 24 hours in anaerobic conditions.
- 3- Prepare serial ten-fold dilutions  $(10^{-1} 10^{-6})$  of the culture using sterile normal saline.
- 4- Transfer 0.1 ml from each of the last three dilutions into sterile Petri dishes, add MRS agar (contain 1% CaCO<sub>3</sub>) into the inoculated plates, mix well and let the plates to solidify.
- 5- Incubate at 37°C for 24- 48 hours in anaerobic conditions.
- 6- Observe the plates and select the colony which shows the clear area surrounding it.
- 7- Pick up such colony; perform gram staining, catalase test and subculture it on similar media in order to diagnosis it later.