

Production of Citric Acid

Citric Acid is the most important organic acid produced by fermentation of carbohydrates, is a colorless, crystalline acid, a solid at room temperature, Melts at 153°C. It is found in fruit juices such as lemon, lime, and pineapple, although many microorganisms can be used to produce citric acid, *A. niger* remains the main industrial producer by utilizing starchy and sugar substrates. Specific strains that are capable of overproducing citric acid have been developed for various types of fermentation processes.

Citric acid applications

*****In the food industry***

- (i) Is the major food acidulated used in the manufacture of jellies, jams, sweets, and soft drinks.
- (ii) Is used for artificial flavoring in various foods including soft drinks.
- (iii) Sodium citrate is employed in processed cheese manufacture.

*****In medicine and pharmacy***

- (iv) Sodium citrate is used in blood transfusion and bacteriology for the prevention of blood clotting.
- (v) The acid is used in effervescent powers which depend for their effervescence on the CO₂ produced from the reaction between citric acid and sodium bicarbonate.
- (vi) Since it is almost universally present in living things, it is rapidly and completely metabolized in the human body and can therefore serve as a source of energy.

***** In the cosmetic industry***

- (vii) Used in astringent lotions such as aftershave lotions because of its low pH.
- (viii) Used in hair rinses and hair and wig setting fluids.

*****Miscellaneous uses in industry***

(ix) In neutral or low pH conditions the acid has a strong tendency to form complexes hence it is widely used in electroplating, leather tanning, and in the removal of iron clogging the pores of the sand face in old oil wells.

(x) Citric acid has recently formed the basis of manufacture of detergents in place of phosphates, because the presence of the latter in effluents gives rise to eutrophication (an increase in nutrients which encourages aquatic flora development).

Citric acid Production

Environmental factors that have been shown to exert an effect on citric acid production are the type and concentration of carbon source of the fermentation medium, nitrogen and phosphate limitations, aeration, trace elements, initial pH and temperature. Besides the regulation of product formation by environmental conditions, strain selection and improvement has become the important factor.

A large number of microorganisms have been employed for citric acid production, but a few of them can produce citric acid in industrial scale. It is reported that *Aspergillus niger* is almost exclusively used for industrial scale production of citric acid, but during the last 30 years the interest of researchers has been attracted by the use of yeasts as citric acid producers.

Easily degradable carbon sources like glucose or sucrose are ideal for citric acid production. For this reason, cane molasses or beet molasses are used. Starch is another source of carbon for citric acid production. For nitrogen source, ammonium ions or urea is added to the medium. Certain trace metals like manganese, iron and zinc are ideal for the growth of *A. niger*. Oxygen is an integral component as *A.niger* is an aerobic organism. The pH of the medium should be less than 3.0 for maximal production of citric acid.

A cost reduction in citric acid production can be achieved by using less expensive substrates, such as apple and grape, carrot waste, orange and pineapple waste, coffee husk, kiwifruit peel, mussel processing wastes, soy residue, rice and wheat bran .

Citric acid is a primary metabolite product and is produced by the involvement of Embden Meyerhof-Parnas (EMP) pathway and TCA cycle. Glucose is first converted to pyruvate. The latter metabolite is decarboxylated to oxaloacetate, which then combines with acetyl CoA to give rise to citric acid. In strains used for citric acid production, TCA cycle is kept active by the involvement of anaplerotic reactions.

Fermentation for Citric Acid Production

It is reported that more than 90% of the citric acid produced in the world is obtained by fermentation. The industrial citric acid production can be carried out in the following ways:

1. Surface process:

This process is the very ancient and is in use in developing countries because it is simple and easy to use. In this process the fungus is grown on the surface of the growth medium, which may be solid or liquid.

A. Solid surface process:

The wheat bran or sweet potato waste adjusted to 70-80% moisture content and pH around 4.0 is sterilized in shallow pans and then sprayed with spores of *A.niger*. Incubation temperature is around 28°C. Sterile air is passed through the incubation chamber for adequate supply of oxygen. Optimal yields of citric acid are produced after 70- 100 hrs of incubation.

B. Liquid surface process:

The liquid fermentation medium basically comprises of beet molasses supplemented with salts and pH adjusted to 4.0 and sterilized. This medium is transferred to a sterile shallow aluminum or steel pans and inoculated with spores of *A.niger* and incubation temperature of 28°C is maintained. Within 24-48 hrs a layer of vegetative mycelium is formed on the surface of medium. Optimal yields of citric acid are produced after a week to 10 days of incubation.

2. Submerged process:

Nearly 80% of citric acid production is carried out by submerged process in stirred tanks of 40-200 m³ or larger airlift fermentors of 200-900 m³ capacity. Submerged fermentation can be carried out in batch, fed batch or continuous systems, although the batch mode is more frequently used. The fermentation vessel is made up of high quality stainless steel as citric acid is very corrosive in nature. The media composition is essentially the same as mentioned in liquid surface process. However, vegetative mycelium and not spore suspension is used for the inoculation of the medium. The vegetative inoculum is prepared by inoculating spores in a small liquid medium. After incubation, the inoculum is sub cultured into a fresh and large volume medium. This process is repeated till desired amount of inoculum in the form of fungal pellets is achieved. Care is also taken to produce pellets which should be small in size (less than 1mm diameter) and have fluffy centers.

Commercially production of citric acid is generally by submerged fermentation of sucrose or molasses using the filamentous fungus *Aspergillus*. During the past twenty years, interest in the use of solid-state fermentation (SSF) as an alternative to submerged fermentation has been increasing. SSF offers numerous advantages, starting with the use of agro-industrial residues. This process has lower energy requirements, produces less wastewater and is environmental - friendly as it resolves the problem of solid wastes disposal.