



## RECENT ADVANCEMENTS IN FERMENTATION

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**Abstract-** Conversation of sugars into organic acid or alcohol depending upon an aerobic and anaerobic process. Naturally, it occurs in many food items and Humans. On increasing the demand for different food products in society, different microorganisms such as bacteria, yeast, and fungi are being used to produce different beneficial products for humans on an Industrial Scale<sup>1</sup>. Different processes, source materials, and different species of fungi and other microorganisms are nowadays being used to produce the desired fermented product.

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**Introduction:** Although fermentation may be defined in many ways broadly it is a process to produce specific products like food products, vaccines, antibodies, etc. with bulk growth of microorganisms. Louis Pasteur made a remarkable contribution in the earlier stage of fermentation. Zymology refers to the study of fermentation.

The history of fermentation starts from the Neolithic era. In that era, people were using the fermentation process to produce food and beverages. The food preservation Industry has a vast demand for lactic acid to preserve pickles, cucumbers, and many other items. It is also used to produce beverages having alcohol such as wine and beers. The process of fermentation also occurs in humans as well as animal's stomach. Since ancient times humans have known about fermentation. They have used it to produce food and beverages. The lactic acid is produced in yogurt and sour food during preservation.

The main organic acid produced nowadays by the fermentation process is citric acid. Although an Islamic alchemist JJ Hayyan in the 8<sup>th</sup> century discovered citric acid it was isolated and crystallized in 1784 by C.W. Scheele from lemon juice. Later in year 1890, the production of citric acid began at the Industrial level. In 1893 C. Wehmer worked on *Penicillium* mold and proved that fungi can produce citric acid. Due to low toxicity, it is used widely as an acidulant in food and pharmaceutical industries. A study shows that the industrial consumption of this acid in food, pharmaceuticals, and other sectors is 70%, 12%, and 18% respectively.<sup>2-3</sup> It is used as a preservative, flavor enhancement, antioxidant, etc. in food, cosmetics, beverages, the pharma industry, and chemical industries.<sup>4</sup> The flavor of food items and drinks such as jams, candies, Jellies, etc. are also enhanced with citric acid.<sup>5</sup> The best way to produce citric acid nowadays is through the state fermentation process.<sup>6</sup>

Citric acids are produced synthetically from glycerol or acetone. Different workers ( Torres et al<sup>7</sup>., 1998; Fernado et al<sup>8</sup>., 2000; Adachi et al<sup>9</sup>.,2003; Haq et al<sup>10</sup>., 2004) have worked on it. Whereas *Aspergillus niger* is the main fungus used for commercial production of citric acid by a submerged fermentation process using sucrose and molasses.

Different workers have worked with different microorganisms ( Kapoor et al<sup>11</sup>; 1983) with bacteria *Bacillus licheniformis*, *Corynebacterium* spp., and *Bacillus subtilis*. Maltey and Allan<sup>12</sup>, 1990 and Kubicek<sup>13</sup>. 1998 worked with different species of fungus *Aspergillus* such as *awamori*, *foetidus* and *Pencillium restrictum*. Yeast such as *candida lipolytica*, *C. intermedia* and

*Saccharomyces cerevisiae* ( Crolla and Kennedy<sup>14</sup>, 2001., Archer et al<sup>15</sup>., 2001; Kamzolova et al<sup>16</sup>, 2003). Out of these the best choice for the production of citric acid is *Aspergillus niger* due to its high yield and ability to ferment with various cheap materials ( Schuster et al<sup>17</sup>., 2002). Cost is one of the factors for any product. To reduce the cost Kiel et al<sup>18</sup>.,1981; Hang and Woodams<sup>19</sup>., 1986; Khare et al<sup>20</sup>., 1995; worked with different agricultural waste such as orange peel, cotton waste, apple, kiwi fruit, etc.

**Materials and Experimentation:-**The general experimental techniques involved Preparation, inoculation, and incubation of different media. In the present work, the analysis involves the unfermented substrate i.e. molasses and citric acid formed along with culture tubes seeding, inoculation of production, and inoculum media, incubation of production, inoculum media, and culture tubes. The different media such as culture, inoculum, and production are sterilized.

Although there are different factors responsible for producing citric acid successfully at minimum cost the most important ones are the selection of fungal strain. The others are the selection of raw materials and cultural conditions.

**Fungal Strains-** The present investigation involves the selection of different fungal strains because different fungal strains are used for different products such as *Aspergillus oryzae* are used to ferment *soyabeans*, rice, and potatoes to produce different products. A distilled Japanese alcoholic beverages called Koji for Sochu are produced by *Aspergillus foetidus*. In certain fruits and vegetables, a disease is found called black mold. This disease is produced by a fungus called *Aspergillus niger*. This *Aspergillus niger* is very efficient to ferment different materials such as molasses to produce citric acid.<sup>21</sup>

The different species of *Aspergillus* are used for citric acid production but the best result were found with *Aspergillus niger* both at laboratory and Industrial scale. Some of the species of *Aspergillus* used in citric acid production are *Aspergillus oryzae*, *Aspergillus flavus*, *Aspergillus wentii*. *Aspergillus foetidus* and *Aspergillus niger* belong to NCIM-647, NCIM-650, NCIM-661, NCIM-511, and NCIM-683 respectively.

In all the fungal strains the NCIM denotes the National Collection of Industrial microbes. These fungal strains may be obtained from the National Chemical Laboratory in Pune, India. The yield percentages for different species differ in the same production medium.

**Fermentation Processes-** There are two types of fermentation processes, one is Submerged and another is solid state fermentation. In the submerged process at the Industrial scale, the bioreactors are operated in batch, fed-batch, or continuous mode to culture different types of microorganisms producing a wide range of products.

In batch cultivation, there is a closed system having medium, nutrients, and Inoculum. At the initial stage, the volume of culture broth remains constant. This process involves several steps such as medium formulation, filling and sterilization of bioreactor, Inoculation, Cultivation, and product harvesting.

Alcoholic beverages like wine, whisky, and rum are widely produced by submerged batch process. At a major scale, citric acids are also produced by this process using molasses as raw material with the help of *Aspergillus niger*.

Other than these there are fed-batch processes which are open systems in which some of the nutrients are gradually added to regulate the cell metabolism. It has an advantage over batch cultivation to regulate the cell metabolism by sequential and precise feeding of nutrients.

The continuous process has an open system where nutrients are continuously added and culture broth is removed at the same time due to which the volume of culture broth is constant. It has many advantages such as there is possibility to set up optimum conditions for maximum and long-term product synthesis. Despite many advantages, there are also certain limitations such as there are possibility of contamination.

In the solid-state fermentation process, the microorganisms are cultured on the surface of the water-insoluble substrate. This process has limitations in the transfer of heat, oxygen, and nutrients due to the growth of microorganisms on the surface of the solid substrate. The heat is generated by microbial metabolism and can reach up to 3000 kcal from 1 kg of the assimilated substrate ( Bellon- Murel et.al 2003)

**Conclusion-** Fermentation fulfills numerous needs of society through the food Industry. There is always a need to increase production and decrease the formation of byproducts. The intensification of bioprocess is possible by engineering aspects, selection of microorganisms, and mutagenesis to improve the production strains. The different fermentation processes have different advantages and limitations. Some of the mutagens of *Aspergillus* species<sup>22</sup> have exciting results in certain conditions and incubation periods. In this

way, there is still further scope to increase the production.

#### References-

1. Leona Paulová, Petra Patáková, and Tomáš Brányik Advanced Fermentation Processes Aug 2013 89-110, <https://www.researchgate.net/publication/259193501>.
2. Pandey A, Soccol CR, Rodriguez-León JA, Nigam P. Production of organic acids by solid-state fermentation. In: Solid-state fermentation in biotechnology –fundamentals and applications. 113-126. Asiatech Publishers. New Delhi (2001).
3. Soccol CR, Prado FC, Vandenberghe LPS, Pandey A. General aspects in citric acid production by submerged and solid-state fermentation. s. 652-664. In: Concise Encyclopedia of Bioresource Technol. The Haworth Press. New York.(2003)
4. Sarangbin S, Krimura K, Usami S. Citric acid production from cellobiose from 2 deoxyglucose- resistant mutant strains of *Aspergillus niger* in semi-solid culture Appl Microbiol Biotechnol. 40:206-210.(1993).
5. Archer DB. Filamentous fungi as microbial cell factories for food use. Current Opinions Biotechnol. 11:478-483. (2000)
6. Adham NZ. Attempts at improving citric acid fermentation by *Aspergillus niger* in beet-molasses medium. Biochemical Technol. 84:97-100. (2002).
7. Torres, N. V., Lopez, J. C., Rivero, M. G. and Rojas, M.G. . Kinetics of growth of *Aspergillus niger* during submerged, agar surface, and solid-state fermentation. Process Biochemistry 33, 103-107. (1998)
8. Fernando, A. V., Carlos, G. A. and Torres, N. V. Metabolism of citric acid production by *Aspergillus niger*.Biotechnology and Bioengineering 70, 82-108. (2000)
9. Adachi, D. M., Toyama, H., Yamada, M., Shingawa, E. and Matsushita, K. New developments in oxidative fermentation. Applied Microbiology and Biotechnology 60, 643-653. (2003)
10. Haq, I., Ali, S., Qadeer, M. A. and Iqbal, J. Citric acid production by mutants of *Aspergillus niger* from cane molasses. Bioresource Technology 93, 125-130 (2004)
11. Kapoor, K.K.,Chaudry, K.,Tauro,P. Citric acid. In: Prescott and Dunn's Industrial Microbiology. Reed,G.(ed.). UK: MacMillan Publishers Ltd. pp. 709-747 (1983)
12. Matthey, M. and Allen, A. Metabolic accumulation in *Aspergillus* species.

- Biochemical society Transaction 18, 1020-265 (1990).
13. Kubicek, C. P. . The role of sugar uptake and channeling for citric acid accumulation by *Aspergillus niger*. Food Technology and Biotechnology 36, 173-175. (1998)
  14. Crolla, A. and Kennedy, K. J. . Optimization of citric acid production from *Candida lipolytica* Y-1095 using n-paraffin. Journal of Biotechnology 89, 27-40. (2001)
  15. Archer, D. B., Mackenzie, A. and Jeenes, D. J. Genetic engineering; Yeasts and filamentous fungi. In: Basic Biotechnology. 2nd edn. Ratledge, C. and Kristiansen, B. (eds.). Cambridge University Press, Cambridge pp. 95-126. (2001)
  16. Kamzolova, S. V., Shishkanova, N. V., Morgunov, I. G. and Finogenova, T. V. . Oxygen requirements for growth and citric acid production of *Yarrowia lipolytica*. Federation of European Microbiological Societies FEMS Yeast Research 3, 217-222. (2003)
  17. Schuster, E., Dunn-Coleman, N., Frisvad, J. C. and Van Dijek, P. W. On the safety of *Aspergillus niger*—A review. Applied Microbiology Biotechnology 59, 426-435.(2002)
  18. Kiel, H., Gurin, R. and Henis, Y. Citric acid fermentation by *Aspergillus niger* on low sugar concentration and cotton waste. Applied Environmental Microbiology 42, 1-4. (1981)
  19. Hang, Y. D. and Woodams, E. E. Microbial production of citric acid by Solid-state fermentation of Kiwi fruit peel. Journal of Food Science 52, 226-227. (1987)
  20. Khare, S. K., Krishna, J. and Gandhi, A. P. Citric acid production from okara (soy-residue) by solidstate fermentation. Bioresource Technology 54, 323-325 (1995).
  21. Samson RA, Houbraken J, Summerbell RC, Flannigan B, Miller JD Common and important species of fungi and actinomycetes in indoor environments. In: Microorganisms in Home and Indoor Work Environments. New York: Taylor & Francis. pp. 287–292. ISBN.(2001).
  22. subedar Yadav, Fungal strains for mycological production of citric acid, International Journal of Pharmaceutical chemistry and analysis, Volume8, Issue2, pp 59-61, April-June 2021.