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# Introductory Chapter: A Brief Overview on Fermentation and Challenges for the Next Future

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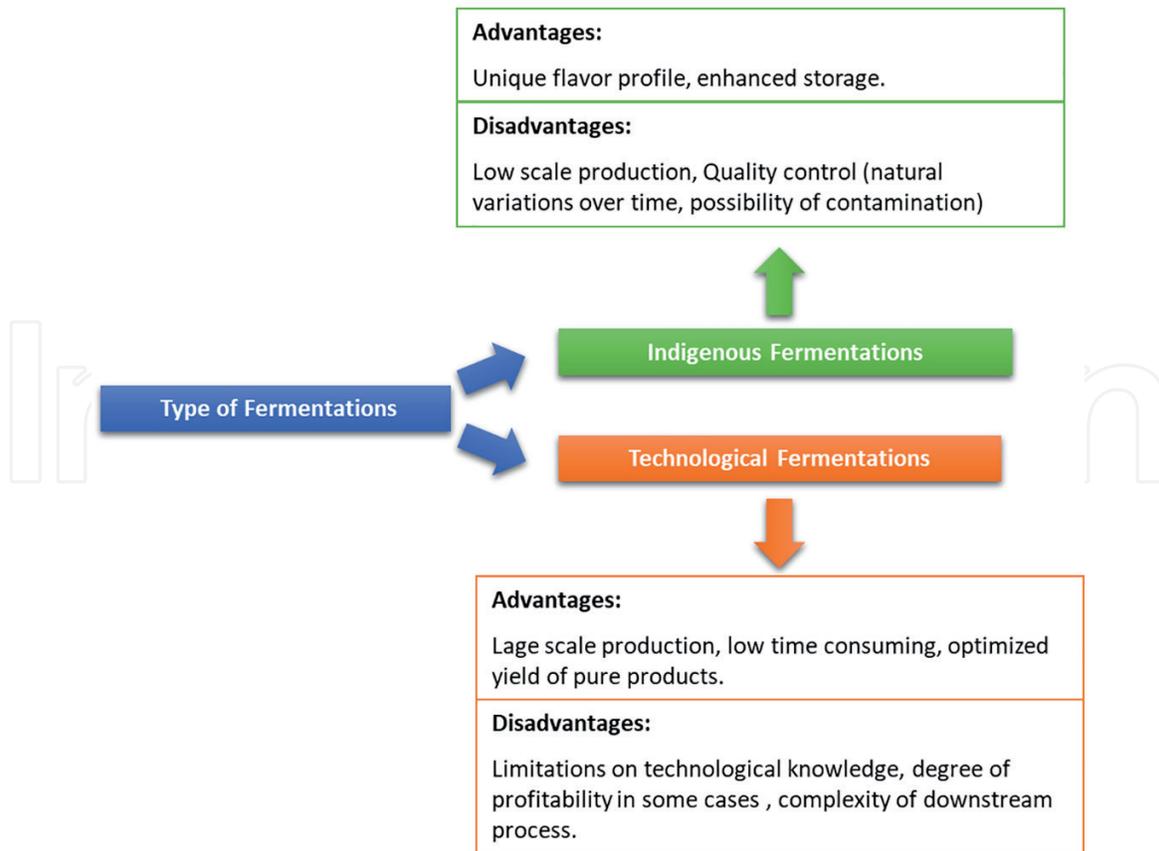
## 1. Introduction

Several authors stated that the word ‘fermentation’ comes from the Latin word *fermentare*, which means ‘to leaven’, while others confirm that it comes from the Latin verb *fervere*, which means ‘to boil’. From these Latin terms, several definitions have been used so far to explain not only the concept of fermentation but also its applications. Thus, the earliest use of the word ‘fermentation’ mostly referred to natural metabolic processes driven by wild and unidentified microbes. Other more recent definitions state that fermentation is a process in which a substance breaks down into a simpler substance usually due to the metabolic activity of microorganisms like yeast or bacteria. In the context of physiology and biochemistry, fermentation processes are usually metabolic pathways producing modifications in organic molecules thanks to the action of microorganisms and/or enzymes [1].

In the strict biochemical sense of the term, fermentation involves the action of anaerobic organisms on organic substrates. However, modern usage extends definition to the microbiological formation of smaller organic molecules, whether aerobic or anaerobic [2]. In applied fields like food or beverage production, fermentation involves any process conducted by microbial activities which brings about a desirable change to a foodstuff or beverage. The component products of fermentation may be isolated from the feedstock and purveyed as pure substances, unlike fermentation of antiquity (e.g. ethanol vs. wine).

From the very beginning, microorganisms were described as the main organisms able to carry out fermentation, but, in general, fermentation is widely distributed in nature. From a biological point of view, fermentation is a way of extracting energy from molecules, and it is one of the common metabolic processes to all living beings: bacteria, archaea and eukaryotes. Thus, fermentation provides ATP thanks to the degradation of organic nutrients (usually under anoxic conditions). In animals for instance, fermentation occurs within the gastrointestinal tracts thanks to microbial flora [1, 3].

Fermentation-based processes have been of great interest for humans since the antiquity due to their potential applications. The origins of fermentation are difficult to track down, and it is assumed that the first fermentation process was discovered accidentally when salt was incorporated with food. Few historians have traced signs of fermentation dating as far back as 7000 BC. At that time, human beings made fermented foods like beer, wine, leavened bread (made primarily by yeasts) and cheeses (made by bacteria and moulds) since Neolithic times in West regions. These societies were soon followed by East Asian regions (6000–1500 BC), where



**Figure 1.** Simple classification of fermentation based on the nature of the process (criteria: processes based on traditional procedures vs. processes based on technological facilities).

fermented foods, yoghurt and other fermented milk products, pickles, sauerkraut, vinegar (soured wine), butter and a host of traditional alcoholic beverages were made following local procedures [4–6].

The aims of using fermentation-based processes have been evolving along time, due to cultural and social issues or due to technology and engineering facilities. Consequently, fermentation has been extensively used in all cultures worldwide to improve food storage for short midterms or to make beverages or local dishes in which fermented sauces, vegetable meat or fish increase the culinary offer. This kind of fermentation is commonly known as ‘indigenous fermentation’/‘classical fermentation’ (it is used to produce foods and beverages using natural microbes following traditional procedures based on cultural practices) (**Figure 1**). Through indigenous fermentation, many products have been standardised and commercialised (ales, natural yeast; cheeses, natural fungi; wines, natural yeast), whilst many other products are made and commercialised at limited quantities for specialised markers or even remain uncommercialised (products of indigenous/local cultures). This is the case of products like kefir, kimchi, sauerkraut, etc. [7, 8].

The results of the evolution of fermentation-based process are made possible through ‘technological fermentations’ (**Figure 1**), which offers several advantages in order to upscale processes.

## 2. Microorganism and enzymes as key tools for fermentation

The nineteenth century was probably one of the periods in which the concept of fermentation as well as its applications focused the attention of professionals from all fields of knowledge. This was mainly due to the French chemist

Louis Pasteur (mid-nineteenth century) who connected, for the first time, yeast metabolic activity to fermentation. Since that time and up to the beginning of the twentieth century, significant number of studies summarised basic descriptions of fermentation processes mainly driven by microorganisms [9–11]. Thus, eukaryotic and prokaryotic microbes able to carry out anaerobic metabolic pathways have been considered good targets for fermentation-based processes so far. During the last four decades, most of the studies focused on fermentation were related to food or beverage production, descriptions of natural fermentations, production of marketed compounds at large scale or technologies to carry out industrial fermentations. Between the descriptions of these natural processes, it is worthy to highlight the following with potential implications in medicine or industries:

- i. The role of microflora in mammals (including human beings and ruminants) [3, 12, 13]
- ii. The production of highly demanded secondary metabolites through fermentation (antibiotics, pigments, prebiotics, etc.) [14–17]
- iii. The production of biomolecules like proteins, amino acids, fatty acids [18], enzymes, vitamins or surface-active compounds [19]

The interest of the use of microbes at industrial scale in fermentation processes was higher at the end of the last century, when some studies reinforced the exploitation at mid or large scale of ‘conventional’ microbes. Shortly after this period, several works increased the interest of scientific community on looking for extremophilic microbes showing ‘rare’ or ‘specific’ metabolic capabilities (even under anoxia) susceptible of being used in industries (Food, Green Biotechnology, Biomedicine, etc.). This was the case of the use of hyperthermophilic microorganisms first [20, 21], members of Archaea domain [22] or nonconventional yeast like *Yarrowia lipolytica* from which production of oils and fats has been recently reported [23].

More recently, other fields of knowledge have evolved thanks to the knowledge of microbial fermentation. Thus, two new areas of research have emerged:

- i. The production of biogas, biodiesel, biofuel and their derivatives [24]
- ii. The design and development of biosorption and bioremediation processes (involving microbes or their isolated enzymes) for soils and wastewater treatments [25]

Not only microorganisms but also their enzymes have contributed significantly to the development of fermentation-based processes. Several processes aim for the fermentative production of microbial enzymes which are further used at large scale mainly in biotechnology [26]. As an example, the use of lignocellulose as a rich and sustainable globally available carbon source to produce bioproducts has only been possible thanks to the use of cellulolytic and hemicellulolytic enzymes produced by different microorganisms including filamentous fungi, yeasts and bacteria [27, 28]. Enzymes from extremophilic microorganisms reveal as promising candidates for industrial processes. The reason beyond this potential use is that most of these enzymes show high catalytic efficiency under some of the extreme conditions used in several processes like high temperature, high ionic strength, extreme pHs or low water availability [29].

### 3. Optimising fermentation processes: impact of technology on fermentation

Technology was coupled to fermentation making possible large-scale production for commercial purposes. Thus, the development of modern engineering, biotechnology and related advanced techniques has connected traditional food fermentations with large-scale production approaches, in which product quality and safety are guaranteed.

To obtain better integrated functions of microbial cells and enzymes, evolutionary engineering combined with other biotechnologies has attracted more attention in recent years. Classical laboratory evolution has not only been proven effective to letting more beneficial mutations occur affecting different genes but also has some inherent limitations such as a long evolutionary period and uncontrolled mutation frequencies [30].

With the arrival of 'genomics era' (genomics, transcriptomics, metagenomics, metabolomics, proteomics, etc.) and 'synthetic biology' approaches, new ways of exploring fermentation are possible due to the possibility of selecting markers and improving cellular transformation strategies [31]. Thanks to these molecular biology approaches, the production of biomolecules through fermentation at large scale is more efficient, low time-consuming and low cost [32].

The development of fermentation technology is still being carried out in all aspects. This is intended to improve the yield and quality of products, reducing the costs of production and looking for processes environmentally friendly. Increasing fermentation products can be done by optimising the factors that influence the process from the aspect of the microbe itself, the environment and the technological facilities. Among these factors, the following have promoted optimization [33]:

- i. Type of feeding of the bioreactor: batch, fed-batch and continuous mode of operation
- ii. Nature of fermentation: one or more than one step; solid vs. liquid
- iii. Type of microbial cells: single strain or mixed culture processes
- iv. Oxygen availability: aerobic, microaerobic and anaerobic processes
- v. Characteristics of inoculation and incubation: ratio of inoculation, agitation rate (to optimise mixing), and continuous control of pH

Other approaches combine microbes and technology at micro-/nanoscale. This is the case of strategies based on electrochemistry, which have been reported as successful approaches, mainly in wastewater and sludge treatments [34].

### 4. Challenges related to fermentation for the next future

As mentioned before, fermentation sustains many processes in food and beverage production at global scale as well as other processes like the production of marketed biocompounds: antibiotics, hormones, pigments, bioplastics, etc. Despite the intensive research efforts on fermentation-based processes, which involve various scientific areas such as plant/microorganism genetics, biochemistry, biomass chemistry and process engineering, the progress of the global use of fermentation has interesting challenges to address in the next future. This is particularly

significant in the case of bioethanol production as a fuel alternative (it is still rather slow compared to the fast-growing demand on such biofuels worldwide).

We are now entering the post-genomic age at a time when many genomes from plants, fungi and microorganisms used in industrial fermentation or microorganisms isolated from food fermentations have already been sequenced. This offers a new knowledge-based approach to the exploitation of these organisms for fermentation related to different industrial activities, from metabolic engineering of microorganisms to produce antimicrobials or nutritional, to the molecular mining of activities yet unknown, but which could benefit food production as well as the production of market biocompounds. Besides, the availability of the genomes of many pathogenic and spoilage bacteria may open new possibilities for the design of novel antibiotics which target essential functions of these problematic bacteria. The real challenge of the genomics and proteomic era, as it applies to food systems, is the harnessing of this wealth of information for improved culture performance and activities, thereby improving the safety and quality and composition of global food supply.

Another important challenge involves technology. Some microorganisms of interest recently described for industrial fermentations require 'extreme conditions' for growth like high salt concentration or highly acidic pHs. This is the case of halophilic or acid thermophilic microbes, respectively. Operating under these conditions causes corrosion (which affects the half-life of most of the bioreactor currently available), thus negatively affecting the implementation of these microorganisms at large scale.

Finally, the design of fermentation processes based on circular economy is still a challenge. Some recent approaches tend to use food wastes as raw materials to design sustainable processes based on acidogenesis, fermentation, methanogenesis, solventogenesis, photosynthesis, oleaginous process, bioelectrogenesis, etc., in order to obtain various products like biofuels, platform chemicals, bioelectricity, biomaterial, biofertilizers, animal feed, etc. which can be utilised for FW valorisation [35].

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## **Conflict of interest**

The author declares no conflict of interest.

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