FOOD FERMENTATION

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Keywords: Acetic acid, Alcohol, Amino acid, Ascomycota, Bacteria, Beer, Bread, Butyric acid, Cacao, Cheese, *Clostridia*, Coffee, Deuteromycota, Eukaryots, Fermentation, Milk, Fungi, Homoacetic bacteria, Industrial fermentation, Lactic bacteria, Metabolism, Microbial ecology, Prokaryots, Propionic acid, Sausage, Tea, Vanilla, Wine, Yeast.

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Summary

Most foods we consume are highly perishable. Their high moisture and nutrient contents create excellent breeding conditions for all types of microorganisms. Some microorganisms can be pathogenic or form toxic compounds, while others can cause direct spoilage of the food. But not all microbial changes in the food are necessarily adverse. Several bacteria, yeasts, and mold strains modify the food systems very fast and the conditions become such that they remain the predominant species. The chemical changes occur mainly under anaerobic conditions through a process called fermentation. Not only are the fermented foods not toxic, they are mostly free of pathogens. The majority show higher digestibility and are richer in nutrients such as vitamins and protein. To successfully obtain high quality fermented food products, it is important to know which microorganisms participate, how they metabolize food components, and how the environmental conditions should be setup for optimum fermentation. Once this is known, it is possible to scale up the fermentation process and to mass-produce fermented food products or fermentation ingredients from fermentable substrates.

1. Introduction

All seem to be going to the same place. It is the end of summer and the tall trees in the forest bear bright yellow fruits. The rains are over and the time of plenty in the region are coming to an end. Birds, monkeys, apes, and other climbing animals, tall mammals such as giraffes and elephants reach the fruit first. A couple of days go by and the remaining fruit is ripe and begins to fall, forming piles. A sweet, fruity aroma starts to permeate the forest and the rest of the animals join the feast. The next day the feast becomes a celebration, and after a while animals of all kinds can be seen stumbling back and forth, most collapsing after a few steps, immediately falling asleep. Nature created wine for the animals to share and most surely *Homo*s took part in this celebration, and wine and other alcoholic beverages have been an essential component of all human cultures ever since.

Perishable foods that were transformed into safe, longer lasting products by the action of fermenting microorganisms immediately became important components of human diets. Be it *sauerkraut* in the west and kimchi in the east, or cheese, yoghurt, *tempeh* from cows milk in the west and goat and camel milk in the east; wine, beer, *mezcal*, *tequila*, *sake*, and many other alcoholic beverages; sausages and fermented fish; all of these products, either produced at home or at an industrial level, have remained important components of the human diet. Some have also had an important nutritional impact because they contain higher levels of vitamins and an improved digestibility. The environment created by fermenting microorganisms is toxic to most pathogenic organisms, making these products safer to eat.

Fermentation has expanded from the transformation of perishable foods to longer-lasting ones, to the utilization of specific components synthesized during the fermentation process. Several chemical compounds can be produced under safer, more economic, and higher yielding conditions, utilizing fermentation instead of chemical synthesis. These compounds are used by different sectors of the industry: food, pharmaceutical, and chemical, etc., and include alcohols, organic acids, vitamins, amino acids, and volatile flavor compounds.

Recently, it was established that a few fermenting microorganisms remain alive in the intestines of humans and other animals and that their presence seems to foster the development of the 'right' intestinal microfauna, thus promoting health. Fermented products containing this type of microfauna have been called probiotics.

As more knowledge on the physiology, metabolism, and ecology of microorganisms becomes available, new applications of their fermentation capabilities will be found. At the same time, only a small fraction of microorganisms have been described, and one can only wonder what the newly described ones will bring with them.

The possibility of expressing one kind of metabolic physiological behavior in microorganisms not originally having such has extended the capabilities of fermentation to a new area, biotechnology.

2. Definitions

The fermentation process has had many definitions. The term directly signifies 'a gentle bubbling or boiling condition' as that seen during wine or beer production. For Louis Pasteur it was 'life in the absence of oxygen'. Many dictionaries call it 'chemical change with effervescence' and 'microbial or enzymatic anaerobic transformations of organic substrates'. Some textbooks have defined it as 'the catabolism of an organic compound to compounds whose average oxidation state is the same as that of the initial substrate but whose total energy content is lower', or 'a metabolic process in which organic compounds serve both as electron donors and electron acceptors'.

A broader definition has been proposed for fermentation, as being 'a process in which chemical changes are brought about in an organic substrate through the action of free enzymes or those present in the microorganisms'. From a biochemical point of view, fermentation is 'a metabolic process in which carbohydrates or related substrates are partially oxidized in the absence of external electron acceptors, and the electron acceptor is an internal organic compound produced during carbohydrate breakdown'.

3. Microbial Ecology

Plant or animal tissues consist of cells whose chemical composition represents an excellent source of nourishment for most organisms on earth - herbivores, carnivores, parasites, and pathogens. Another group of microorganisms, the decay microbes, has developed evolutionary physiologic and metabolic adaptations to use plant or animal tissues as their ecological niche, and to degrade the tissues to simple chemical compounds. The initial decay microbe populations are represented by fermenting microorganisms that modify the chemical composition or the decaying tissues in such a way, by increasing the levels of alcohols and organic acids, that they out-compete other microorganisms for prolonged periods. The fermenting populations are followed by other microbial populations until all tissues are degraded mainly to water, ammonia, and carbon dioxide.

The fermenting, decaying microfauna make no distinction between dead animals or plant tissues or perishable foods. Fortunately, most fermented tissues acquire an aroma and flavor that is palatable to many animals, including humans. Some fermented food products suffer partial putrefaction but retain an acceptable flavor. Under natural conditions, the diversity of the decaying microfauna is immense and completely dependent on biotic and abiotic environmental parameters.

3.1 The Fermenting Microorganisms

All human cultures have developed a wide variety of fermented foods and have become accustomed to a consistent level of quality in these products. Consistent quality can only be achieved through the use of consistent raw materials, fermenting microorganisms, and processing conditions. Thus, knowledge of the physiology and metabolism of the different fermenting microorganisms has become vital.

The diversity of fermenting microorganisms is immense, and the knowledge base about their taxonomy, physiology, and metabolism is widely dispersed, and in most cases, each aspect is treated separately.

KINGDOM	DIVISION	PHYLUM	CHARACTERISTICS / EXAMPLES
Archaea Anaerobes		**Euryarchaeota**	Non-spore forming
		Methanogens	Gram positive /negative; *Methanococcus, Methanosarcina*
		Halophiles	Gram negative; *Halobacterium, Natronobacterium*
		Crenarcheota	Thermoacidophils; Thermoproteaceae; Desulphurococcaceae
Eubacteria Diverse metabolism	**Gracilicutes** *Gram negative*	**Proteobacteria**	Purple non-sulphur bacteria; non-oxygenic phototrophs . - **Group:** *Acetobacter, Gluconobacter*; *Caulobacter*; *Hyphomonas, Bradyrhizobium, Rhizobiaceae, Rhodobacter, Azospirillum, Rickettsia, Erythrobacter* . - **Group:** *Alcaligenes,Bordetella; Nitrosomonas; Leptothrix, Thiomonas; Gallionella, Hydrogenophilus, Massilia timonae, Methylobacillus, Chromobacterium, Neisseria, Azospira, Ferribacterium, Spirillum* . - **Group:** *Escherichia, Salmonella, Coxiella, Legionella, Methylococcus, Pasteurella, Haemophilus, Azotobacter, Beggiatoa, Vibrio, Xanthomonas, Xylella* . - **Group:** *Bdellovibrio, Desulfomonas, Desulfovibrio, Geobacter, Lawsonia* . - **Group:** *Campylobacter, Dehalospirillum, Helicobacter, Sulphurospirillum*
		Spirochaeta	Anaerobes; heterotrophs *Spirochaeta, Borrelia, Treponema*
		Cyanobacteria	Oxygenic phototrophs *Nostoc, Anabaena, Prochloron, Lyngbya*
		Saprospirae	Gliders; *Bacteroides, Flavobacterium, Cytophaga*
		Chloroflexa	Filamentous, green, non-sulphur, phototrophic, gliding; *Chloroflexus*
		Chlorobia	Anoxygenic, green, non-sulphur, phototrophic; *Chlorobium*
	Tenericutes Wall-less	**Aphragma- bacteria**	*Mycoplasma, Spiroplasma*
	Firmicutes Gram positive; protein walled	**Endospora**	Endospores; *Bacillus, Chlostridium, Listeria,Streptococcus, Staphylococcus*
		Pirellulae	Heterotrophs; some obligate parasites); *Chlamydia, Planctomyces*
		Actinobacteria	High-Guanine/Citosine; *Corynebacterium, Streptomyces*
		Deinococci	Radio-resistant; *Deinococcus, Sarcina*
		Thermotogae	Fermenters; *Thermosipho, Thermotoga*

Table 1: Taxonomy of bacteria (Superkingdom Prokarya)
[From: Benson D.A., Karsch-Mizrachi I., Lipman D.J., Ostell J., Rapp B.A., and Wheeler D.L. (2000). *GenBank*. Nucleic Acids Res. **28 (1)**, 15-18.]

KINGDOM*	PHYLUM	CHARACTERISTICS
Protoctista	**Myxomycota** **Plasmodiophoromycota** **Dictyosteliomycota** **Acrasiomycota**	• True slime molds; plasmodium changes with appearance of a fungus • Endoparasites with sporangiogenous and cystogenous plamodia • Cellular slime molds, pseudoplasmodium • Cellular or communal slime molds, pseudoplasmodium
Chromista	**Sagenista** **Hyphochytriomycota** **Oomycota**	• Basal chromists, heterotrophic; net-plasmodium; non-fermenting • Aquatic chytrid-like organisms; some parasitic; chitin cell wall; non-fermenting • Water molds (*Saprolegnia*) and downymildews (*Phytophthora*) - filamentous protists; some fermenting
Fungi	**Chytridiomycota**	• *Asexual spores - zoospores; absorptive nutrition, few plant pathogens* *Class* Chytridiomycetes; *Orders* Blastocladiales, Chytridiales, Monoblepharidales
	Zygomycota	• Asexual reproduction in sporangia or conidia; sexual spores diploid, zygospores *Class* Zygomycetes – *Orders* Entomophthorales (pathogenic), Endogonales (mycorrhizal), Mucorales (pathogenic, fermentative)
	Ascomycota	• Sexual spores ascospores in an ascus *Class* Ascomycetes *Subclass* Hemiascomycetidae – *Order* Taphrinales (*Taphrina*), *Subclass* Hymenoascomycetidae [Discomycetes] – *Order* Helotiales (*Sclerotinia*, *Monilinia*); [Pyrenomycetes] – *Orders* Xylariales (*Glomerella*), Diaporthales (*Diaporthe*), Erysiphales (*Erysiphe*), Hypocreales (*Giberella*) *Subclass* Loculoascomycetidae – *Orders* Dothideales (*Cercospora*), Pleosporales (*Setosphaeria*) *Subclass* Plectomycetidae – *Orders* Microascales (*Ceratocystis*), Onygenales (*Trichophyton*)
	Basidiomycota	• Mushrooms, shelf fungi, puffballs, stinkhorns, bird's nest fungi, jelly fungi, and the plant pathogens rusts and smuts; saprophytes, plant symbionts, phytopathogenic; sexual spores basidiospores on a basidium; dikaryon a free-living vegetative stage; non-fermenting *Class* Basidiomycetes *Subclass* Holobasidiomycetidae - *Orders* Aphyllophorales (*Ganoderma*), Exobasidiales (*Exobasidium*), Agaricales (*Agaricus*, *Boletus*, *Pleurotus*) *Subclass* Teliomycetidae – *Orders* Uredinales (*Puccinia*), Ustilaginales (*Ustilago*)

	Deuteromycota	• Fungi Imperfecti, presumably Ascomycota; sexual spores unknown *Class* Deuteromycetes *Subclass* Blastomycetidae (pathogenic) – *Order* Cryptococcales, (*Candida*) *Subclass* Coelomycetidae (phytopathogenic) – *Orders* Melanconiales (*Colletotrichum*), Sphaeropsidales (*Phoma*) *Subclass* Hyphomycetidae (phytopathogenic) – *Orders* Moniliales (*Fusarium*, *Alternaria*), Agonomycetales (*Sclerotium*)

* Monera, Plantae, and Animalia, contain no groups of organisms recognized as fungi.

Table 2: Key to Kingdoms containing taxa, traditionally considered Fungi.
[From: Benson D.A., Karsch-Mizrachi I., Lipman D.J., Ostell J., Rapp B.A., and Wheeler D.L. (2000). *GenBank*. Nucleic Acids Res. **28 (1)**, 15-18.]

Taxonomic knowledge can be used to identify how closely related organisms are, and profound knowledge on one organism can be used to deduce the characteristics of closely related, but lesser described organisms. Biologists have developed a common language to refer to all organisms - systematics. Phylogenetic systematics is the way biologists reconstruct the pattern of events that have led to the distribution and diversity of life. Classification is based on the evolutionary history of life, such that it can be used to predict properties of newly discovered or poorly known living or fossil organisms.

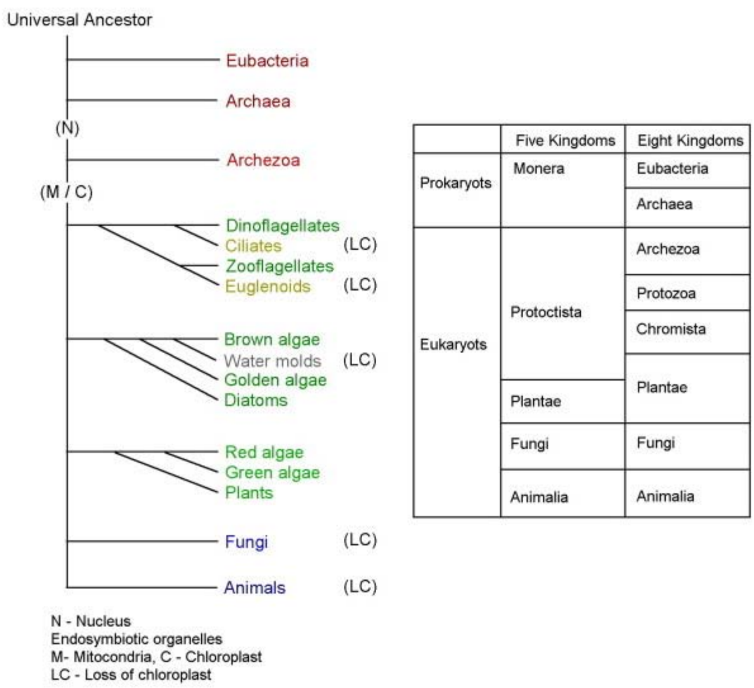

Figure 1: Hypothetical phylogeny of living organisms.
[From: Margulis L. and Schwartz K.V. (1988). *Five Kingdoms,* pp. 376. New York: W.H. Freeman & Co.]

A proposed phylogenetic tree showing the grouping of organisms in a Five Kingdom

System is shown in Figure 1, accompanied by a more recently proposed Eight Kingdom System. Fermenting organisms can be found in three of the known Kingdoms: Eubacteria among the Prokaryots, and Chromista and Fungi among the Eukaryots (Tables 1 and 2).

3.2 Prokaryots and Eukaryots

Prokaryots are single-celled organisms lacking the complex internal membrane systems (nuclear membrane, organelles, endoplasmic reticulum, etc.) present in eukaryots. Prokaryots are metabolically diverse while eukaryots retain only part of the metabolism of prokaryots. Prokaryots have one chromosome and reproduce mainly asexually, while eukaryots have multiple and more complex chromosomes and reproduce mainly sexually.

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Bibliography

Barnes R.S.K. (1988). The Diversity of Living Organisms. Malden, MA: Blackwell Science Ltd. [This book gives important information on the taxonomy and evolution of microorganisms]

Benson D.A., Karsch-Mizrachi I., Lipman D.J., Ostell J., Rapp B.A., and Wheeler D.L. (2000). GenBank. Nucleic Acids Res. 28 (1), 15-18. [A publication that proposes the use of genetic information for the taxonomy of organisms. Finer adjustments on specific classifications of microorganisms can be achieved with information on their genomes]

Cavalier-Smith T. (1998). A Revised Six Kingdom System of Life. Biol. Rev. Camb. Philos. Soc. 73, 203-266. [Margulis' book on the 'Five Kingdoms' has been an important reference on evolution since its publication and this publication suggests important adjustments to it]

Crueger W. and Crueger A. (1989). Biotechnology: A Textbook of Industrial Microbiology, Second Edition, 357 pp. Sunderland, MA: Sinauer Associates, Inc. [This is a classic book on the subject. Very thorough on microbiological aspects and on fermented products]
Margulis L. and Schwartz K.V. (1988). Five Kingdoms, 376 pp. New York: W.H. Freeman & Co. [A classic text since its publication and it is an indispensable source of information on the evolution of all organisms]

National Center for Biotechnology Information Taxonomy. Homepage of the NCBI
http://www.ncbi.nlm.nih.gov/Taxonomy/taxonomyhome.html. [A very important source with updated information on microbial taxonomy]

Pederson C.S. (1979). Microbiology of Food Fermentations, Second Edition. Westport, Connecticut: AVI Publishing Co., Inc. [Another classic book on food biotechnology that encompasses almost all fermented products available throughout the world and the microorganisms involved in their production]

Prescott S.C. and Dunn C.G. (1959). Industrial Microbiology. New York: McGraw-Hill Book Company. [One of the first text books dealing with the whole process in the manufacture of fermented products, from the microorganisms involved to the raw materials, the processes, and the final product. A text that transformed industrial fermentations into an interdisciplinary discipline]

Wheeler D.L., Chappey C., Lash A.E., Leipe D.D., Madden T.L., Schuler G.D., Tatusova T.A., and Rapp B.A. (2000). Database resources of the National Center for Biotechnology Information. Nucleic Acids Res. 28 (1), 10-14. [An important database on many aspects of microorganisms employed in industrial fermentations]

Websites

Website http://www.ncbi.nlm.nih.gov/Taxonomy/taxonomyhome.html [Homepage of the NCBI *National Center for Biotechnology Information* Taxonomy Browser]

Biographical Sketches

Pedro Wesche-Ebeling is Head of the Department of Chemistry and Biology at the Universidad de las Américas – Puebla, México. Professor Wesche-Ebeling received his M.S. and Ph.D. degrees in Food Science at Oregon State University; he is a member of the Institute of Food Technologists and the Society of Economic Botany. Professor Wesche-Ebeling's teaching interests include food chemistry, biochemistry, and economic botany areas. His research interests involve the study of the potential of phyto-chemicals as substitutes for artificial additives in foods (pigments, antioxidants, antimicrobials, etc.), as well as of the nutraceutical potential of these compounds. This research focuses on Mexican native plants, in an effort to transform some into important commercial crops, and therefore increase the incomes of farmers and steer an interest in the preservation of biodiversity.

Jorge Welti-Chanes was born in Puebla City, Mexico; he obtained his B.S. in Biochemical Engineering (1976) and Master of Science in Food Engineering (1979) at the Instituto Tecnologico y de Estudios Superiores de Monterrey (ITESM), Mexico. In 1979, he obtained his Ph.D. in Chemical Sciences within the Food Technology area from the Universidad de Valencia, Spain. Dr. Welti-Chanes has been Professor at ITESM, and since 1977, at the University of the Américas-Puebla (UDLA). At UDLA, he has taught at undergraduate and graduate levels in the Department of Chemical and Food Engineering and in the Department of Chemistry and Biology. He was Head of the department for one year, and then Dean of the Engineering School (1986-1988). Since 1989, he has been Academic Vice-President of UDLA as well as Professor and Researcher at the same institution. Dr. Welti-Chanes' research focus is mainly on food drying, water activity, minimal processing of fruits, process simulation, and emerging technologies. He has been coordinator of diverse international research projects within the mentioned areas. He was World President of the International Association of Food and Engineering (1997-2000). Dr. Welti-Chanes is also author of more than 90 scientific publications in refereed journals and more than 150 presentations in international congresses. He is coeditor of the books, *Food Preservation by Moisture Control; Inventario de Alimentos de Humedad Intermedia Tradicionales de Iberoamérica; Engineering and Food for the 21st Century;* and *Harvesting, Handling, and Preservation of Fruits and Vegetables by Combined Methods at Rural and Village Levels* (Technical Manual of FAO). He is a member of Editorial Boards for different journals and book series. He evaluates research projects in Argentina, Chile, Uruguay, Venezuela, and Mexico. He is advisor to different food enterprises in areas such as processing, quality assurance, and optimization of processes. Also, Dr. Welti-Chanes has advised more than 20 universities in Mexico and Latin America on Strategic Planning of Educational Processes.