Ecology of Fermented Foods

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Abstract

Fermented foods arise in the human relationship to the microbial environment. Human survival is connected to yeasts and bacteria that produce lactic acid and alcohol in preserved foods. This constitutes a fermentation ecosystem that embodies the succession of species, partitioning of resources, disturbance and equilibrium found in larger ecosystems. Fermented foods are preserved by microbes that live in food storage vessels. In many societies, the contribution of fermented food has been central. Fermentation ecosystems can be used as an engaging instructional tool to illustrate ecological concepts and lead to a more complex understanding of the ecology of human nutrition.

Keywords: *alcohol, ecosystem, fermentation, food, lactic acid*

Introduction: Fermentation as an Ecological Process

The word ecology conjures images of the outdoors, and discussions of human food tend to focus on things we can see with the naked eye. We would like to highlight the importance of the indoor, microbial ecology of fermented foods, such as beer, cheese, bread, yogurt, and other foods which constitute microbial ecosystems that live in human households across the globe. In this paper we describe fermentation as a human ecological process, and suggest that fermentation is a wonderful vehicle for teaching ecological concepts.

Food fermentation is practiced by human cultures all over the world. It is a major component of human survival in places where preserved food is a necessity. Production of fermented food does not require knowledge of the biologically-mediated nature of fermentation, because the biota that carry out fermentation are present worldwide. Fermentation organisms are ambient in the environment, whether humans make use of them or not. The relationships embodied in preserved foods can be treated as microbial ecosystems, sitting on countertops and stored in food cellars around the world.

The main components of the fermentation ecosystem include: microbes (yeast and bacteria), organic material to be fermented, a solution in which the fermentation takes place, a vessel with a controlled gate, and various tools which may be used to develop and monitor the fermentation (thermometer, hydrometer, siphoning tubes, etc.). This is an ecosystem in that it is a complex of living and non-living components that are viewed in terms of their interactions in a specific place. Beer biochemistry has recently been used to teach food chemistry (Pelter 2006), but the emphasis in fermentation studies is usually on non-living chemical *components*. In this essay, we will treat the chemical pathways of microbial metabolism as a black box, in order to shift attention toward the interactions within the microbial *environment* (see Figure 1).

Knowledge of microbial metabolism is not necessary for the production of fermented foods, and indeed wasn't even proposed until 150 years ago (Pasteur 1858). Yet evidence of controlled fermentations has been found at the earliest agricultural settlements in China, the Middle East and South America (McGovern et al. 2004; Moseley et al. 2005; Guasch-Jané et al. 2005). Today, all human cultures rely on fermented food products and the microbes that produce them. Fermentation is not only a component of human ecology, but also has an ecology of its own. Over the past 150 years, microbiologists have uncovered this unseen world of ecosystems that have always lived in our food. This essay will introduce food fermentation as a human ecological process, with a focus on pickled foods, alcoholic beverages, microbial evolution and domestication.

Fermented Foods

Fermentation is a natural process that unavoidably affects the human food supply worldwide. Wild fermentation bacteria and yeast cover the continents and permeate ecosystems, in the air, soil, water, and guts of animals; they are a natural resource available to people all over the world. Although fermentation is everywhere, it is rarely understood. Fermentation is the microbially mediated conversion of sugars into products such as alcohol and lactic acid. When consumed by humans, fermented foods often introduce microflora that inhabit the human body (Ross et al. 2002; Reid et al. 2003; Picard et al. 2005). Many common foods contain fermentation microorganisms and their by-products (see Table 1). In this article, the word “fermentation” will be used simply to refer to the process by which alcohol or lactic acid are produced by living cells in solutions that contain sugars (Ribéreau-Gayon et al. 2000).

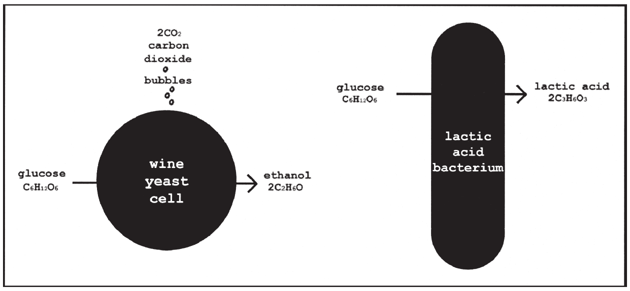

Figure 1. In order to focus attention to inter-special relationships, and relationships to the fermentation substrate, microbial metabolism is treated as a black box in this essay. Alcoholic fermentation carried on by yeast (left) and lactic acid fermentation carried on by bacteria (right) are central to the fermentation of food and drink. Note: yeast and bacteria cells come in different sizes and shapes; this figure is not an illustration of scale.

There are two kingdoms of life in fermentation ecosystems: fungi and bacteria. Fermentation fungi include yeasts (such as *Saccharomyces*), which produce alcoholic beverages and molds (such as those found in bleu cheese). Fermentation bacteria are responsible for pickles, cheese, and cured sausages. Generally, yeasts are used to produce ethanol, and bacteria are used to produce lactic acid. There are exceptions: bread yeast is employed to produce carbon dioxide bubbles in leavened dough, and *Gluconobacter* is a genus of bacteria that can produce acetic acid (vinegar).

Fermented foods are generally produced using plant or animal ingredients in combination with fungi or bacteria which are either sourced from the environment, or carefully kept in cultures maintained by humans. Just as living organisms cover the surface of the earth, fermentation microbes cover the surface of the organisms. Wild yeasts are found living on grapes (Chamberlain et al. 1997), and bacteria line the human digestive tract. Kimchi, a spicy pickled food that Koreans eat at almost every meal, has scores, perhaps hundreds of species living in it (Lee et al. 2005), and previously unknown species of bacteria are being discovered in Kimchi microflora (Kim et al. 2003; Lee et al. 2002; Kim et al. 2000; Yoon et al. 2000). The ecology of fermented foods is still a frontier of discovery.

Table 1. Some common fermented foods. The fermentation organisms listed comprise only some of the most well known players in the fermentation ecosystem. There are many more.

Food	Raw Material	Fermentors (selected species)
Beer	Grain malt	*Saccharomyces cerevisiae*
Bread	Grain flour	*Saccharomyces cerevisiae*
Butter	Milk	*Streptococcus* spp.
		Leuconostoc spp.
Cheese	Milk	*Lactobacillus* spp. (primary)
		Lactococcus spp. (primary)
		Pediococcus spp. (primary)
		Streptococcus spp. (primary)
		Leuconostoc spp. (secondary)
		Propionibacter spp. (secondary)
Chocolate	Cacao bean	*Saccharomyces cerevisiae*
		Candida rugosa
		Kluyveromyces marxianus
Coffee	Coffee bean	*Erwinia dissolvens*
Kefir	Milk	*Saccharomyces kefir*
		Torula kefir
Kimchi	Napa, Daikon	*Lactobacillus plantarum*
		Lactobacillus brevis
		Streptococcus faecalis
		Leuconostoc mesenteroides
		Pediococcus pentosaceus
Miso	Soy bean	*Aspergillus oryzae*
Olives	Olives	*Candida spp.*
		Cryptococcus spp.
		Debaryomyces hanseii
		Lactobacillus spp.
		Saccharomyces spp.
Pickles	Cucumber	*Leuconostoc mesenteroides*
		Lactobacillus spp.
Sauerkraut	Cabbage	*Coliform spp.*
		Leuconostoc plantarum
		Lactobacillus spp.
Sausages	Meat and spices	*Pediococcus spp.*
		Lactobacillus spp.
Soy Sauce	Soy bean	*Aspergillus oryzae*
Tempeh	Soy bean	*Rhyzopus oligosporus*
Vinegar	Fruit juice	*Saccharomyces cerevisae*
		Gluconobacter spp.
		Acetobacter spp.
Wine	Grapes	*Saccharomyces cerevisiae*
		Saccharomyces bayanus
Yogurt	Milk	*Lactobacillus bulgaricus*
		Streptococcus thermophilus

Ecosystem Succession in Pickled Foods

One can explore basic ecosystem processes in a kitchen or a classroom by making sauerkraut. Making sauerkraut involves a bacterial ecosystem succession in which each population of bacteria create the conditions for the following bacteria to thrive. Making sauerkraut is simple: 1) chop cabbage, 2) layer with salt, 3) weight down the cabbage and salt in a container (see Figure 2). Call this the "disturbance phase" of the sauerkraut ecosystem. Three tablespoons of salt is sufficient to pull the juices out of a gallon of chopped cabbage via osmosis. The salt must not contain iodine or other anti-caking agents. The vegetable juices mix with the salt to create a brine which inhibits the growth of putrefying organisms while creating prime conditions for the growth of *Coliform* bacteria, which are ubiquitous in the air. Call *Coliform* the "pioneer species" of the sauerkraut ecosystem. *Coliform* bacteria produce acids, lower the pH of the kraut, and set the stage for *Leuconostoc* bacteria to colonize the medium. The *Leuconostoc* bacteria lower the pH of the sauerkraut further still, thus creating the conditions for *Lactobacillus* to grow (Katz 2003). *Lactobacillus* adds the characteristic taste of lactic acid to the food, a smooth note in the otherwise tangy kraut. Lactic acid can create a temporary equilibrium state, which is the ultimate goal of food preservation via fermentation. Refrigeration halts the fermentation process. If left out in room temperature air, the kraut will get more acidic and sour, eventually reaching a point of inedibility. This phenomenon is similar to the aging of mineral soils, which lose organic matter and become more acidic as the get older.

Another way to explore the concept of ecological succession is to make cheese. Dairy fermentations involve the conversion of lactose to lactic acid through several successional phases. Raw milk, after it is collected, contains multiple bacterial populations. Fresh milk has a neutral pH and a warm temperature — perfect conditions for the rapid multiplication of *Lactococcus* bacteria. *Lactococcus* immediately begins producing lactic acid, lowering pH, and thus creating favorable conditions for the growth of *Lactobacillus* bacteria, which convert lactose to lactic acid and lower the pH further still (Flórez and Mayo 2006). By using up nutrients, lowering pH, and producing metabolic wastes, each microbe helps create conditions favorable for its successor. Many cheeses are also "surface ripened" by molds, yeast, or other bacteria that significantly alter the flavor. Some yeasts may follow *Lactobacillus* and consume lactic acid. This is the end stage for some live-rind cheeses, whence the fermentation is halted (Marcellino and Benson 1992). The site-specific adaptation of wild microbes can be quite strong: in one study, when cheeses were innoculated with human-selected bacterial strains, the cheeses were actually ripened via a sequence of wild microflora ambient in the environment, who crowded out the introduced bacteria and rendered the innoculum superfluous (Brennan et al. 2002).

Figure 2. The components of lactic acid fermentation (sauerkraut): vegetable (cabbage), non-iodized salt, weight, and time.

Cheese recipes stress the importance of controlled temperature, water content versus salt level, and storage location, which are effectively climate, moisture regime and geography of the dairy fermentation ecosystem. For centuries, cheese-makers have refined their methods of guiding the succession of bacteria and yeast that produce the best cheeses, by creating controlled conditions in which selection pressures determine microbe survival (Marcellino et al. 2001). Although the cheese-makers may have simply thought of themselves as altering temperature, water, and salt (and indeed they were), they were also altering microbial population growth dynamics, ecosystem species composition, and microbial survival, so that certain flavor compounds were produced in a stable manner. As such, this point of equilibrium was the ecological stopping point in the cheese succession — under proper conditions, a stable cheese can last for weeks, months, or even years, making the nutrition-value of the dairy product a longer- lasting alternative than it would be without the biological transformation.

The same equation roughly applies for all pickled foods and hard sausages: lactic acid fermentation generally involves reducing the amount of water in the food, increasing the amount of salt, and controlling temperature, such that the desired microorganisms lower pH and ultimately produce lactic acid, effectively preserving the food while producing otherwise unavailable flavors and nutritional compounds. Most lactic acid fermentations also produce a salty liquid solution (e.g. brine, whey, tamari), which also may be considered a biologically active fermented food. This is in contrast to alcoholic fermentations in which the final product is usually dead.

Alcoholic Fermentation as a Natural Resource

Alcoholic fermentation ecosystems are dominated by one genus of yeast: *Saccharomyces*. Beers are produced by fermenting maltose from malted grains. Wines are produced by fermenting sugary fruit juices. Species of *Saccharomyces* are ambient in the environment, and are present on the skins of fruits such as ripe grapes (Holloway et al. 1990). Old world grape wines were products of the native yeasts in their regional ecosystems. Modern oenology has shown that there is a succession of fermenting microorganisms, that yeast strain populations differ between batches and years, and wine quality is therefore vineyard and vintage dependent (Schutz and Gafner 1994). The wine yeasts can out-compete the pathogenic onslaught introduced during barefoot stomping without pasteurization, and produce a 12% alcohol (and thus pathogen-free) wine for the table. Domesticated *Saccaromyces* (store-bought wine yeast, brewers yeast, or bread yeast) combined with different ingredients and environmental conditions, can be manipulated using simple equipment (see Figure 3) in a pedagogical setting to demonstrate environmental chemistry, population dynamics, nutrient limitation, and competition for finite resources.

Figure 3. The components of wine fermentation: fruit (elderberry), pot, bucket, carboy, siphon tube, and airlocks to allow carbon dioxide to escape from the containers.

The components of a finished wine or beer include flavor compounds that also function as non-living chemical components of the fermentation environment. Controlled alcoholic fermentation involves pasteurizing a sugary solution called a must, and then later adding selected strains of *Saccaromyces* to the cooled must to convert the sugar to alcohol. The different tastes of wines, ports, meads, beers, and distilled liquors comes from the different relative proportions of alcohol and the other compounds remaining in the final product, as a consequence of must preparation and yeast-strain selection. Distinguishable flavor compounds include (but are not limited to) organic acids (acetic acid, citric acid, tartaric acid, malic acid, lactic acid), esters, carbonyl compounds (diacetyl, aldehydes), sulfur compounds (dimethyl sulfide, hydrogen sulfide), and residual sugars (Berry and Slaughter 2003). This is the environmental chemistry of the must, and there are several books that focus primarily on the chemistry aspects of alcoholic fermentation in relation to yeast activity (Janson 1996; Ribéreau-Gayon et al. 2000).

Experimental brewing is an application of Liebig's law of minimum. In nature *Saccharomyces* are limited by the unavailability of sugars. Home-scale alcoholic fermentations are often started by producing a "starter culture" in a plastic bottle consisting of sugar, fruit juice, and yeast. Adding a packet of wine yeast, beer yeast, or even bread yeast should work, because they are all the same species (though bread yeast strains will not produce delicious beverages). The yeast multiplies rapidly and begins releasing carbon dioxide (CO_2) and ethanol within a few hours. The CO_2 must be released periodically or else the pressure will build up and burst the bottle. The goal of making a starter culture is to produce a large colony of live *Saccharomyces* to inoculate a must. The starter culture and the must both go through a climax stage known as high krausen when the maximum quantity of yeast cells is achieved. The goal of making a starter culture is to add it into the must during high krausen.

To demonstrate Liebig's law of minimum, an otherwise difficult to illustrate environmental chemistry concept, alcoholic fermentations may be shown to be limited by the absence of other nutrients. *Saccharomyces* cannot live on sugar alone. To illustrate, prepare a one gallon must of sugar dissolved in water, and add wine yeast to ferment the sugar. After a short flurry of activity, sugar water fermentations become stagnant or "stuck," as the microbial ecosystem is nutrient-limited. It may be possible to overcome stuck sugar water ferments by adding nutrients: a cup of fruit juice should suffice. One must be careful, however, not to add juice that contains potassium sorbate or excessive sulfites. These compounds are added to prevent commercial fruit juices from fermenting, and they will have the same effect on a must.

Fruits and grains have evolved mechanisms to prevent fermentation from taking place, and yeasts have evolved mechanisms to help them compete for sugars in short supply. For instance, there is a class of yeasts known as "killer yeasts." Killer yeasts are more competitive than other *Saccharomyces* strains; they use extra-cellular enzymes to kill other yeast strains and then proceed to scavenge the dead yeast for food. Since there is no primary production in the fermentation ecosystem, whatever carbon is added at the beginning is the maximum carbon that will be available for the duration of the fermentation (assuming no new ingredients are added). The yeasts are respiring CO_2 out of the system, so it should not be surprising that some strains of yeast have evolved a tendency to predate other yeasts in order to win the competition for finite carbon. With unpasteurized musts, killer yeasts may be added to ensure speedy fermentations, as the killer yeasts also tend to work very quickly.

In the absence of primary producers, the fermentation ecosystem is thus an ecosystem of consumers. Nutrient cycling, habitat/niche, and competition for resources may be demonstrated by manipulating the availability of nutrients provided at the outset by the humans who set up the system. Even without the killer yeasts, there is evident cycling of nutrients from dead yeast cells (sediment) that settle at the bottom of the solution. There are different fermentation "climates": ale yeasts prefer to ferment at 70˚F, whereas lager yeasts do better around 55˚F. *Rhyzopus oligosporus* (a mold fungus) can't ferment soybeans to tempeh unless it is very warm (>80˚F). The pods of coffee, chocolate, and vanilla beans are fermented outdoors in the sun in tropical parts of the world, and likely provide unique habitat to microbes that fulfill that role. In the absence of oxygen, yeasts (*Saccharomyces*) ferment grape juice to wine, but in the presence of oxygen, acetic acid bacteria (*Acetobacter*) can oxidize the ethanol to acetic acid (vinegar) (Fleet 1992). These are the micro-environmental conditions of the fermentation food web.

Discussion

The concept of a *whole ecosystem* is unpopular in some circles and many have abandoned the idea that ecosystems have boundaries. For those who maintain that discrete ecosystems are useful, fermentation projects are an excellent illustration. A fermented food is easier to explain than a large ecosystem such as an archipelago, and it is more immediately useful as well. Environmental education might do well to add a few fermentation projects to its repertory, even when the intention is to show the lack of autonomy of open biotic communities. San Francisco Sourdough cultures taken to other parts of the world lose their San Francisco flavor qualities after about six months, so a baker producing San Francisco Sourdough bread in New Jersey will have to constantly import new starter cultures to maintain the integrity of a San Francisco Sourdough bread operation. Tasting the fermented foods at different points in time allows for a primary experience of the link between flavor and an evolving biotic community of yeast populations. Where else do humans get to experience the tangible results of a biotic community changing over millions of generations in a short period of time?

The repeated fermentation of a single food product in a given place over long periods of time is a process of domestication. Registered appellations (the official wine-growing regions of a country) are a product of local yeasts, local soils, and local cave conditions that produce distinct regional wines, even when different regions are using the same grape cultivar. The same may be said for cheeses. For centuries, cheese-makers have domesticated the yeast strains that produce the best cheeses by creating controlled conditions in which human domestic selection pressures determine yeast survival (Marcellino et al. 2001). By altering salt, water, and temperature, cheese-makers were altering microbial environments, affecting microbial survival based on adaptation to the cheese-making processes over centuries. The influence of human choice over fermentation processes reveals different ecosystem responses to these perturbations.

Indeed some fermented food cultures are themselves biodiverse, comprised of symbiotically associated bacteria and fungi. Kefir is a fermented milk product in which lactose has been converted to alcohol and lactic acid. Kefir is produced by kefir "grains," which are tapioca-like colonies of bacteria and yeast (see Figure 4). Recent studies have found kefir grains to include populations of *Zygosaccharomyces, Candida, Leuconostoc, Lactococcus, Lactobacillus*, and

Table 2. Some *how to* books for first-time fermentors.

Title	Authors	Publisher	Year
And That's How You Make Cheese	Sokol, S.	iUniverse	2001
Classic Sourdoughs: A Home Baker's Handbook	Wood, E.	Ten Speed Press	2001
Home Cheese Making	Carroll, R.	Storey Books	2002
The Compleat Meadmaker	Schramm, K.	Brewers Publications	2003
The Book of Miso	Shurtleff, W.	Ten Speed Press	2001
The Joy of Home Brewing, 3rd ed.	Papazian, C	Collins	2003
The Joy of Home Winemaking	Garey, T.	Collins	1996
The Permaculture Book of Ferment and Human Nutrition	Mollison, B.	Tagari Publications	1993
Wild Fermentation	Katz, S.	Chelsea Green Publications	2003

Cryptococcus, which grow differentially in successional phases (Witthuhn et al. 2005). Adding kefir grains to milk produces a sour yogurt-like beverage within about 48 hours, and if the solution is sealed during fermentation it will develop effervescence (Katz 2003). Complete kefir fermentation produces a lactose-free food that lactose-intolerant people can digest. Kefir is being investigated as a leavener for bread (Plessas et al. 2005), a starter culture for cheeses (Goncu and Alpkent 2005), and a source of kefiran, an insoluble polysaccharide with antibacterial and cicatrizing properties that show potential for use in medicine (Rodrigues et al. 2005). Kefir is a stable probiotic food that may be kept for months without spoiling, and the kefir colonies provide a tangible, visible sense of the biodiversity that inhabit this fermented food. Kefir points to a means of solving environmental problems by using diversity instead of eliminating it.

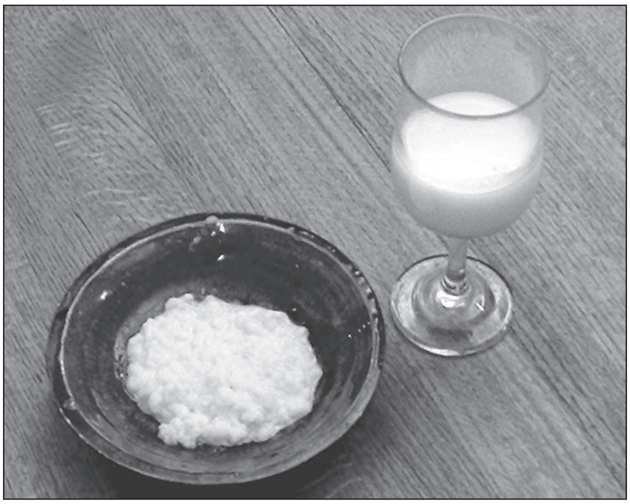

Figure 4. Kefir "grains" and kefir beverage. Kefir combines many of the properties of various fermented foods in a single project.

The ecology of fermented food can be considered as both a simple model and a domain of complex experimentation. Fermented foods can be used to construct ecosystem demonstrations that are small-scale and short-term. Fermenting foods can be as easy as following a recipe (see Table 2), and the illustration of distinct ecosystem concepts can be achieved by modifying one variable at a time. It is difficult to capture the essence of continental ecosystems, but it is easy to distinguish the boundary of a fermentation container, and to observe the processes taking place within those boundaries, on a microbiological scale. These processes include, but are not limited to: effect of temperature/climate on species composition, effect of species composition on environmental chemistry, ecological succession and population dynamics such as competition, climax, and equilibrium, the concept of nutrient limitation, selection pressures, domestication, and biodiversity. The deliberate use of fermented foods shows how all life processes are inextricably linked with ecological processes, and that the patterns of culture which last the longest may be those that mimic the dynamics of ecosystems.

Endnotes

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