### Unit III

### Fermented fruits & vegetables products (Sauerkraut, Cucumber & Olives)

Wherever vegetables are grown and consumed, it is almost certain that fermented versions exist. Moreover, despite the wide diversity of vegetables produced around the world, the principles involved in the manufacture of fermented vegetables are very near the same. Like other fermented foods, readily observed variations certainly exist, and are based on aesthetic preferences, as well as the types of raw materials available in particular regions.

The manufacture of fermented vegetables most likely evolved from simply dry-salting or brining vegetables. Salting vegetables was a common means of food preservation and was practiced for thousands of years in Europe, the Middle East, and Asia, and for several centuries in the Americas. In general, salt or brine was added to the fresh raw material as a preservation aid, and then the mixture was packed into suitable containers and stored at an ambient temperature. If the salt concentration was not too high, this practice would have established ideal conditions for growth of naturally-occurring lactic acid bacteria. The ensuing fermentation would have not only enhanced preservation, but it would have also created highly desirable flavor and aroma characteristics.

### **Products and Consumption**

In the United States, there are essentially only three fermented vegetable products that are produced and consumed on a large scale basis. These include sauerkraut, pickles, and olives. The raw materials for these products-cabbage, cucumbers, and olives—are high moisture foods, with little protein or fat (except for olives), and just enough fermentable carbohydrate to support a fermentation (Table 7.1).

Vegetable	$H_20$	Carbohydrate	Protein	Fat
Cabbage	92% - 94%	5% - 6%	1%	_
Cucumbers	95%	2% - 3 %	1%	_
Olives	78% - 80%	2% - 4%	1% - 2%	12% - 14%

Table 7.1. Composition of substrates used in vegetable fermentations.

Other fermented vegetables, such as peppers, cauliflower, and green tomatoes, are also produced, but these are not nearly as popular (at least in the West). There are also many acidified or pickled vegetable products that are made by adding mixtures of vinegar, salt, and flavoring materials to fresh vegetables.

### 1. Sauerkraut

Few fermented foods are produced in such a seemingly simple process as is sauerkraut (Figure 7.1). Only two ingredients, cabbage and salt, are necessary, and once these ingredients are properly mixed, there is little that the manufacturer needs to do until the fermentation is completed. The simplicity of the process is reflected by the U.S. Standards, which states that sauerkraut is the "product of characteristic acid flavor, obtained by the full fermentation, chiefly lactic, of properly prepared and shredded cabbage in the presence of not less than 2 percent nor

more than 3 percent of salt."After fermentation, sauerkraut should contain not less than 1.5% acid (expressed as lactic acid).

The manufacture of sauerkraut starts with the selection of the raw substrate material. Although various cabbage cultivars exist, white cabbage is typically used because it has a mild, slightly sweet flavor and contains 5% or more fermentable sugars (mostly equimolar amounts of glucose and fructose, with a small amount of sucrose). Cabbage used to make sauerkraut should be fully mature, and should contain few outer leaves. Some manufacturers allow the cabbage heads to wilt for a day or two.



Harris, 1998.

# Shredding and salting

Once the outer leaves and any spoiled leaves are removed, the cabbage heads are washed and the core is drilled out. The cabbage (along with the core) is shredded (according to the manufacturers specifications) to make a slaw. The shredded leaves are then weighed and conveyed directly to tanks or are deposited first into tubs or carts and then transferred into tanks. Salt can be added as the slaw is conveyed or it can be added to the slaw when it arrives in the tanks. In either case, both the amount of salt added and the means by which mixing and distribution occur are critical. Usually, between 2% and 2.5% salt is added (by weight), although 2.25% is generally considered to be the optimum. Problems are almost certain to occur if too much or not enough salt is added

or if the salt is not uniformly distributed, because salt performs several essential functions during the sauerkraut fermentation. Very soon after the salt is mixed with the shredded cabbage, water begins to diffuse out from the interior of the plant tissue to the exterior medium, due to simple osmosis. The brine that forms also contains sugars and other dissolved nutrients that diffuse out with the water. Thus, it is this water phase that ultimately serves as the location for most of the microbial activity.

Next, salt (dissolved in the brine) provides the selective conditions that discourage growth of most of the non-lactic microorganisms that would otherwise compete with the lactic microflora. Although salt at a concentration of only 2.25% is, by itself, not ordinarily sufficient to inhibit all of the indigenous, non-lactic bacteria, it is enough to provide the lactic acid bacteria with a substantial growth advantage. Furthermore, combined with other environmental factors, the selective effects of this relatively moderate salt concentration can be increased appreciably (discussed below). Moreover, once the pH has been decreased by the production of organic acids, the combination of salt plus acid contributes significantly to the long preservation properties of the finished product. Finally, salt imparts a desirable flavor to the product and helps to maintain a crisp texture by preventing softening of the tissues.

#### Mixing

The shredded and salted cabbage is then placed into tanks and mixed well to distribute the salt. As noted above, mixing is an important step, because localized regions within the rather heterogenous material may contain more or less than the 2.25% salt that was added to the bulk mixture. Within those pockets, therefore, it is entirely possible that the salt concentration may vary considerably, perhaps by as much as 0.1%. This may result in either too little or too much inhibitory control over the organisms that reside in that microenvironment. If spoilage organisms were able to grow, their products (e.g., slime, pigments, off-flavors) could accumulate and, when the sauerkraut is mixed prior to packaging, contaminate the entire batch of product. It is worth noting that high salt levels can promote spoilage just as readily as low salt levels. For example, the "pink" defect is caused by growth of salt-tolerant yeasts that ordinarily would be suppressed by lactic acid bacteria whose growth is impaired at high salt levels.

The sauerkraut fermentation was traditionally performed in wooden barrels. Wood-stave tanks are still used; however, concrete vats are now common. The latter are lined with fiberglass or plastic, and can hold as much as 50,000 Kg. The cabbage is covered with a plastic, tarp-like material, large enough to drape over the sides of the tank. Water (or brine) is then placed on top to weigh down the cabbage and to drive out and exclude air. This also reduces exposure to airborne organisms, foreign matter, and insects. The weight further enhances formation of a brine, which soon completely covers the shredded cabbage.

### **Fermentation**

The sauerkraut fermentation has long been the subject of interest among food microbiologists as well as microbial ecologists. In fact, many of the biochemical and

microbiological details of the sauerkraut fermentation were described as long ago as the 1930s. This interest has undoubtedly been due, in large part, to the very nature of the fermentation process, in that it involves several different naturally-occurring microorganisms acting as part of a complex ecosystem. Recent reports suggest that bacteriophages may also play an important role in the microbial ecology of the sauerkraut fermentation.

The manufacture of sauerkraut and many other fermented vegetables depends on a succession of organisms that are naturally present in the raw material. Some appear early on in the fermentation, perform a particular function, and then, for all practical purposes, disappear from the product. Other organisms, in contrast, emerge later in the fermentation and then remain at moderate to high levels throughout the duration of the fermentation and post-fermentation process. However, growth of those organisms that occur late in the process depends on those organisms that had grown earlier and that had established the correct environmental conditions. Microbial activity begins as soon as a brine has formed. Initially, the atmosphere is aerobic, with redox potentials (or Eh values) of over 200 mV. However, the combined effects of physical exclusion of air and residual respiration and oxygen consumption by plant cells quickly reduce the Eh and make the environment anaerobic. Thus, pseudomonads, fungi, and other obligate aerobic microorganisms that may initially be present at high levels, have little opportunity for growth. Some of these organisms are also salt-sensitive, further reducing their ability to grow in this environment. Still, at the temperatures used during the sauerkraut fermentation (20°C to 25°C), many other indigenous salt-tolerant, mesophilic, facultative organisms might be expected to grow, including Enterobacter, E. coli, Erwinia, and other coliforms. Instead, these organisms persist for only a short time, perhaps as little as a few hours, due to competition by lactic acid bacteria and the inhibitory effects of the acids produced by these bacteria. The lactic fermentation in sauerkraut occurs in a series of overlapping stages or sequences. These stages and the succession of microorganisms associated with each stage have been very well studied. Remarkably, the fermentation almost always follows the exact same pattern.

The first stage, variously referred to as the initiation or heterolactic or gaseous phase, is marked by growth of *Leuconostoc mesenteroides*. This organism is salt-tolerant and has a relatively short lag phase and high growth rate at low temperatures ( $15^{\circ}C$  to  $18^{\circ}C$ ). Importantly, it metabolizes sugars via the heterofermentative pathway, yielding lactic and acetic acids, CO2, and ethanol. The acidic environment (0.6% to 0.8%, as lactic acid) created by growth of *L. mesenteroides* not only inhibits non-lactic competitors, but it also favors other lactic acid bacteria. The production of CO2 also contributes to making the environment even more anaerobic (as low as \_200 mV), which again favors the more anaerobic lactic acid bacteria. Eventually, however, as the acid concentration approaches 1.0%, *L. mesenteroides* is, itself, inhibited, and within four to six days, this organism is barely detectable.

In the next stage or primary, homolactic, or non-gaseous phase, the decrease in the *Leuconostoc* population coincides with the succession of several other lactic acid bacteria, most notably *Lactobacillus plantarum* and, to a lesser extent, *Lactobacillus brevis*. Although *L. plantarum* is a facultative heterofermentor (meaning it has the metabolic capacity to ferment

different sugars via homoor heterofermentative pathways) and *L. brevis* is an obligate heterofermentor, both organisms are strong acid producers, nearly doubling the acid content to about 1.4% to 1.6%. They are also quite stable in this acidic environment and dominate the fermentation during this period (especially *L. plantarum*). It is not unusual, however, for other lactic acid bacteria, including *Pediococcus pentosaceus*, *Pediococcus acidilactici*, *Lactobacillus cuvatus*, and *Enterococcus sp.*, to be present during the primary fermentation. Finally, as the acidity approaches 1.6% and the pH decreases below 4.0, only the acid-tolerant *L. plantarum* is able to grow. The entire process can take up to one to two months, and the fermentation is generally considered complete when the acidity is at about 1.7%, with a pH of 3.4 to 3.6.

### Packaging and processing

In the United States, commercial products are usually thermally processed, much like other high-acid foods at about 75°C, prior to packaging in cans or jars. Such products are essentially commercially sterile and are stable at room temperature. There is also a market for non-pasteurized, refrigerated sauerkraut that is packaged in glass jars or sealed plastic bags (polybags). These products also have a long shelf-life, provided antimycotic agents, such as benzoate and sulfite salts, are added and the product is kept cold.

### 2. Cucumber

In a very general sense, pickles refer to any vegetable (or fruit) that is preserved by salt or acid. Certainly, the vegetable most often associated with pickles is the cucumber. Currently, about half of the total cucumber crop (1 billion Kg) in the United States is used for pickles. Pickles and pickling technology have a long and rich history. The cucumber was brought from India to the Middle East about 4,000 years ago, and cured versions were eaten at least 3,000 years ago. Cleopatra endorsed pickle consumption, claiming they were responsible (in part) for her beauty. The ancient Roman historian and natural scientist Pliny the Elder and his contemporaries, Roman emperors Julius Caesar and Tiberius, were all fond of pickles. Columbus introduced pickles to the Americas, eventually leading to the origins of a pickle industry on the Lower East Side of New York City. American founding fathers George Washington, John Adams, and Thomas Jefferson reportedly derived inspiration from the pickle. In the United States, pickles are generally divided into three different groups, based on their means of manufacture. Fresh-packed pickles are simply cucumbers that are packed in jars, covered with vinegar and other flavorings, then pasteurized by heat. They have a long shelf-life, even at room temperature. Fresh packed pickles are crisp, mildly acidic, and are the most popular.

Refrigerated pickles are also made by packing cucumbers jars with vinegar and various flavorings, but they are not heated. Instead these pickles are refrigerated, giving them a crisp, crunchy texture and bright green color. Although a slight fermentation may occur, refrigerated pickles have a shorter shelf-life than fresh-packed pickles. Sodium benzoate is usually added as a

preservative. The manufacture of both fresh-packed and refrigerated-style pickles is fast and easy and require few steps (Figure 7.4).



Figure 7-4. Non-fermented pickles and their manufacture. Adapted from Harris, 1998.

# Manufacture of fermented pickles

The actual process steps used for the manufacture of fermented pickles are similar to those used for making sauerkraut. Both rely on salt, oxygen exclusion, and anaerobiosis to provide the appropriate environmental conditions necessary to select for growth of naturallyoccurring lactic acid bacteria. There are, however, several differences between pickle and sauerkraut fermentations. First, salt concentrations are higher than those used for sauerkraut, resulting in the development of a less diverse microflora. In addition, a brine, rather than dry salt, is used for pickle fermentations.Finally,the pickle fermentation process, unlike sauerkraut, is amenable to the use of pure starter cultures and a more controlled fermentation. Indeed, such cultures are now available and some (but not many) pickle manufacturers have adopted controlled fermentation processes.

The manufacture of fermented pickles starts with selection and sorting of cucumbers (Figure 7.5). Only small or immature cucumbers, harvested when they are green and firm, are used for pickles. They are then washed, sorted, and transferred to tanks, and a brine solution is added. The brine typically contains at least 5% salt (or about 20° salometer, where 100° salometer = 26% salt). Because the cucumbers-to-brine ratio is nearly 1:1, the actual salt concentration is actually less. For so-called salt stock pickles, which may be held in bulk for long periods, the initial brine may contain 7% to 8% salt, which is followed by the addition of more salt to raise the total salt concentration to above 12%. For genuine dill-type pickles, the brine

concentration is usually between 7.5% and 8.5%. Dill weed is also added, usually in the seed or oil form. Care must be taken when weighing down the pickles, because the buoyancy of the cucumbers may cause those at the top to become damaged. The large tanks used by large pickle manufacturers are usually located outdoors, and temperatures may, therefore, vary between 15°C to 30°C. The lower the temperature, the longer it takes to complete the fermentation. Thus, in Michigan (the largest northern producer of pickles), fermentation may require up to two months, whereas in North Carolina (the main southern producer), fermentations may be complete in three weeks. At the end of the fermentation, the pH will be about 3.5, with acidities between 0.6% and 1.2% (as lactic).



Figure 7–5. Manufacture of fermented pickles. Adapted from Harris, 1998.

# Pickle fermentation

As noted above, the high salt concentrations used in pickle manufacturing cause the fermentation to proceed quite differently from that in sauerkraut. Only those pickles made using brines at less than 5% salt will allow for growth of *L. mesenteroides*. Although heterofermentative fermentations may promote more diverse flavor development, the formation

of CO2 is undesirable, because it may lead to bloater or floater defects (see below). Moreover, low salt brines may also permit growth of unwanted members of the natural flora, including coliforms, *Bacillus, Pseudomonas,* and *Flavobacterium*. At salt concentrations between 5% and 8%, growth of *Leuconostoc* is inhibited and instead the fermentation is initiated by *Pediococcus* sp. and *L. plantarum*. Pickle fermentation brines typically contain high concentrations of salt and organic acids and have a pH less than 4.5. These conditions are especially inhibitory to coliforms, pseudomonads, bacilli, clostridia, and other non-lactic acid bacteria that would otherwise cause flavor and texture problems. This environment, in fact, is hard even on lactic acid bacteria. However, the latter have evolved sophisticated physiological systems that enable them to survive under very uncomfortable circumstances.

After fermentation, salt stock pickles can be held indefinitely in the brine. However, these pickles cannot be eaten directly, but rather must be de-salted by transfer to water. After several changes (a process called refreshing), the salt concentration is reduced to about 4%. They are then used primarily for relishes and other processed pickle products.

### 3. Olives

Olives refer not only to the usually salty, acidic product known as table olives, but to the fruit from which they are made. The main use of raw olives, in fact, is as olive oil—more than 90% of the total worldwide olive production is used for oil and only 7% to 10% are consumed as table olives. Currently, about 90% to 95% of the olives grown in California are used in the manufacture of table olives (making California among the leaders in table olive production). Although fermented olives are common in Europe and other olive-producing regions, most of the table olives produced and consumed in the United States are not fermented. In fact, more than 70% of the U.S. olive market consists of olives that are simply brined and canned (hence, this type is referred to as California-style olives).

### Manufacture of fermented olives

There are three main styles or types of table olives, based on their method of production (Figure 7–7). Spanish-style (or green Spanishstyle) olives are treated with sodium hydroxide (lye) and fermented. Greek-style or naturally black, ripe-style olives are not treated with lye, but are fermented. The fermentation for both types is mediated by the natural microflora, much like that for other fermented vegetables (discussed below). The third type of olive is the ripe black- or green-style. They are lye-treated, but are not fermented. They may also undergo a special aeration treatment that promotes oxidation of pigments and conversion of a green color to black. This is the type referred to as California-style olives.



Figure 7-7. Types of olives and their manufacture. Adapted from Harris, 1998 and Romero et. al., 2004.

#### Spanish-style

Spanish-style olives are harvested when the skin color is green or straw-yellow. They are then treated with a lye solution for four to twelve hours at 15°C to 20°C to de-bitterize the olives via hydrolysis of oleuropein. The lye concentration may range from 0.5% to 3.5%, depending on the size and type of olive. Once the lye has penetrated to just outside the pit (about two-thirds of the way from the skin to the center), the olives are washed in one to three rinse cycles of water to remove the lye. The pH of the olives after washing should be less than 8.0. Although there should be little residual lye remaining with the olives, a slight amount of bitterness may still be present, which is characteristic of these olives. Following the washing, the olives are moved to tanks or barrels and a brine of varying salt concentrations is added. For some olives, 10% to 15% salt brines are used (giving an actual concentration of 6% to 9%), whereas others start with lower salt brines (5% to 6%), and salt is added later to give comparable final concentrations. Glucose may be added to restore sugars lost during lye treatment and washing steps. The brined olives are subsequently held at 22°C to 26°C. Although the endogenous microbial population is reduced by the lye and washing treatments, the olive production environment still contains a wide assortment of microorganisms. There is, in fact, an opportunity for growth of coliforms, Pseudomonas, Bacillus, Clostridium and other Gram negative and Gram positive bacteria during the first two to four days, especially when low salt brines are used, and before a lactic fermentation can begin in earnest. This so-called first stage of the fermentation is then followed

by a second stage, initiated primarily by *L. mesenteroides* and *Pediococcus* sp. Once the pH nears 5.0, non-lactics are inhibited, and the brine consists only of lactic acid bacteria. After two to three weeks, *L. plantarum*, *L. brevis, Lactobacillus fermentum*, and other lactobacilli displace the leuconostocs, and eventually *L. plantarum* dominates the third stage of the fermentation. The appearance of facultative yeasts at the end stage of the fermentation, however, is not uncommon, nor undesirable, since they may produce ethanol, acetaldehyde, and other flavor compounds. In contrast, growth of *Propionibacterium acnes* is to be avoided, since this organism can raise the pH by fermenting lactic acid.

When the fermentation is complete, the final pH after three to four weeks (or longer for barrel-fermented olives) should be within a range of 3.5 to 4.2, with a titratable acidity (as lactic) of 0.8 to 1.0. Following the fermentation, Spanish-style olives can be pitted, pitted and stuffed (e.g., with pimentos), or packed directly into jars. Pasteurization to promote extended shelf-life is optional.

### Greek-style

There are several significant differences between Greek-style and other olive types. First, these olives are naturally black when they are harvested, in contrast to California-style black olives that rely on oxidation to generate black pigments. Second, Greek-style olives are not lye-treated, giving them a more bitter flavor. Third, the fermentation is mediated not just by lactic acid bacteria, but also by yeast, non-lactic acid bacteria, and even fungi. Some of these non-lactic organisms (e.g., *Pseudomonas* and Enterobacteriaceae) may actually remain in the brine, albeit at low levels, for several weeks before they begin to decline. Thus, the fermentation end-products include not just lactic acid, but also acetic acid, citric acid, malic acid, CO2, and ethanol. This mixed fermentation may result in a less acidic product with a final pH as high as 4.5 and an acidity less than 0.6%. Lower brine concentrations (5% to 10%) may contribute to the more diverse flora that develop in these olives.

Another type of fermented olive that is made in a very similar manner (i.e., no lyetreatment) is the Sicilian olive. The main difference between these olives and the Greek olives is that the Sicilian olives are green (like Spanish olives). Also, while lactobacilli are the main bacteria involved in the fermentation, the dominant species appears to be *Lactobacillus casei*, an organism not ordinarily associated with fermented olives.

### *Ripe- or California-style*

California-style olives are the most popular olives consumed in the United States. Both the black and green versions are produced from the same starting material: green olives (with a bit of cherry-red blush). For both types, the olives are lye-treated, as described above for Spanish olives, except that several applications are used. In the case of green olives, the lye is removed and the olives are washed in water, and then dilute brine is added. The olives are then canned (after a pitting step, if desired) and thermally processed as a low-acid canned food. For black olives, the lye-treated olives are heavily aerated between the lye applications to promote darkening reactions. Aeration can be accomplished either manually, by stirring, or mechanically, by direct injection of air into the tanks. The latter method is preferred because it is faster and reduces opportunities for spoilage. Air, or more specifically, oxygen, promotes a chemical oxidation reaction in which phenolic compounds are polymerized to form first brown, then black pigments. During air exposure, the concentration of two phenols, hydroxytyrosol and caffeic acid in particular, decreases, as the black color increases. Ionic iron can form complexes with the oxidized phenols; thus, iron salts are usually added to these olives to fix the color and prevent fading.