Fermented and Acidified Vegetables

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51.1 INTRODUCTION

Vegetables may be preserved by fermentation, direct acidification, or a combination of these along with other processing conditions and additives to yield products that are referred to as pickles. Pasteurization and refrigeration are used to assure stability of certain of these products. Organic acids and salt (sodium chloride) are primary preservatives for most types of pickles. Lactic acid is produced naturally in fermented products. Acetic acid (or vinegar) is the usual acid added to pasteurized, unfermented (fresh-pack) pickles. Acetic acid also is added to many products made from fermented (salt-stock) cucumbers. Other preservatives such as sodium benzoate, potassium sorbate, and sulfur dioxide may be added to finished products. Although the term "pickles" in the United States generally refers to pickled cucumbers, the term is used herein in a broader sense to refer to all vegetables that are preserved by fermentation or direct acidification. Cucumbers, cabbage, olives, and peppers account for the largest volume of vegetables and fruits commercially pickled. Lesser quantities of onions, tomatoes, cauliflower, carrots, melon rinds, okra, artichokes, beans, and other produce also are pickled.

The fermentation of vegetables is due primarily to the lactic acid bacteria, although yeasts and other microorganisms may be involved, depending on the salt concentration and other factors. Salt serves two primary roles in the preservation of fermented vegetables: It influences the type and extent of microbial activity, and it helps prevent softening of the vegetable tissue. Some vegetables are brined at such high salt concentrations as to greatly retard or preclude fermentation. Salt may be added in the dry form, as with cabbage, or as a brine solution, as with most other vegetables. The concentration of salt used varies widely among vegetables, depending on tendency of the vegetable to soften during brine storage. Softening of brined cucumbers can be reduced or prevented by adjusting the level of salt to inhibit pectinolytic enzymes.[3,4] Fermentation is an economical means for temporary preservation of produce such as cucumbers, cabbage, and olives. The produce is fermented and stored in large tanks until it is needed for further processing. After removal from brine storage, brined cucumbers may be desalted if needed before being finished into various products such as dills, sweets, sours, hamburger dill chips, mixed vegetables, and relishes.[50] Finished salt-stock dill cucumber pickles contain a minimum of 0.6% lactic acid, according to USDA grade standards.[137] The products may or may not be pasteurized, depending on the addition of sugar and other preservatives. Extensive reviews are available on the brining and fermentation of cabbage,[118,131] cucumbers,[48,53] and olives.[52,68,139,140,141]

Direct acidification with acetic acid (without pasteurization) has been a primary method for many years of preserving various pickles and sauces in the United Kingdom, where the products are referred to as acetic acid preserves. British researchers have determined that the minimum acetic acid concentration necessary to achieve satisfactory preservation of all pickles and sauces is 3.6%, calculated as a percentage of the volatile constituents of the product.[9] The high concentration of acid needed for preservation results in such a strong acid flavor, however, that the relative importance of this method of preservation has diminished. Milder acidic flavors are more in demand today, and use of acidification in combination with pasteurization has become more important. Nevertheless, some specialty products such as hot pepper sauce and sliced peppers still are preserved principally by high concentrations of acetic acid without pasteurization.

Fresh-pack cucumber pickles are preserved by mild acidification (0.5% to 1.1% acetic acid[137]) of fresh cucumbers, followed by heating to an internal product temperature of 74°C and holding for 15 minutes, according to the original recommendations of Etchells et al.[30,37,109] Such products are effectively pasteurized, since they are heated enough to inactivate microbial vegetative cells, and sufficient acid has been added to prevent outgrowth of bacterial spores. Although some packers still use this heat process, others now vary the times and temperatures, depending on product type and risk factors (for spoilage problems of non-health significance) acceptable to the packer. Fermented pickles, such as whole genuine dills and hamburger dill chips, may or may not be heated. If pasteurized, these products may be given a milder heat treatment than fresh-pack pickles, such as an internal product temperature of 71°C with no holding time. The fresh-pack process has been applied to peppers and other vegetables. Fresh-pack pickles are considered acidified foods for regulatory purposes. According to the U.S. Food and Drug Administration (FDA), "'Acidified foods' means low-acid foods to which acid(s) or acid food(s) are added; these foods include, but are not limited to, beans, cucumbers, cabbage, artichokes, cauliflower, puddings, peppers, tropical fruits, and fish, singly or in any combination. They have a water activity (a_w) greater than 0.85 and have

a finished equilibrium pH of 4.6 or below. These foods may be called, or may purport to be, 'pickles' or 'pickled'...."[136]

Refrigerated pickles may or may not be fermented before refrigeration. Also, they may or may not be acidified, although mild acidification is highly recommended.[49] Most commercially prepared and distributed refrigerated pickles sold today are not fermented, but are acidified and contain a preservative such as sodium benzoate.

Increasing environmental concerns related to waste disposal are influencing methods for preservation of pickled vegetables, particularly those involving use of salt for bulk storage. The U.S. Environmental Protection Agency (EPA) has proposed a maximum of 230 ppm of chloride in fresh waters,[29] a limit that may not be readily achievable by many vegetable briners who discharge chloride wastes into freshwater streams. Organic acids (lactic and acetic) in combination with calcium chloride and preservatives (e.g., sodium benzoate) are now used instead of sodium chloride for bulk storage of olives for "green-ripe" processing into canned black olives in California.[142] Salt is still used, however, for fermented olives. The use of calcium salts (chloride or acetate) has led to reduced levels of sodium chloride for bulk fermentation and storage of cucumbers. Calcium salts have been found to enhance firmness retention of cucumbers at reduced concentrations of sodium chloride.[14,61,65,84,135] Studies have revealed, however, that spoilage microorganisms may present a serious problem in fermented cucumbers if the salt concentration is too low.[68] Recently, the use of sulfite was proposed as a way to store cucumbers in the absence of salt, and the sulfite removed by reaction with hydrogen peroxide after storage and before conversion into finished products.[100]

51.2 NORMAL FLORA

Fresh produce contains a varied epiphytic microflora (Chapter 50). Pickling cucumbers were found to contain as high as 5.3×10^7 total aerobes, 1.9×10^4 aerobic spores, 9.8×10^5 total anaerobes, 5.4×10^2 anaerobic spores, 6.1×10^6 coliforms, 5.1×10^4 total acid formers, 4.6×10^3 molds, and 6.6×10^3 yeasts per g of fresh cucumber.[46] The numbers increased during storage at higher temperatures (21°C) and humidity (>70% relative humidity). Although some investigators have held that the interior of sound, fresh cucumbers is sterile, others have found microorganisms, mostly gram-negative rods, within the healthy fruit.[106,127] In cucumbers, bacteria were more often near the skin and less often in the central core; in tomatoes, their frequency was highest near the stem-scar and central core and decreased toward the skin.[127] Cabbage contains the greatest number of bacteria on the outer leaves and lower numbers toward the center of the head.[118]

The floral changes during natural fermentation of brined vegetables may be characterized into four stages: initiation, primary fermentation, secondary fermentation, and post-fermentation.[53] During initiation, the various gram-positive and gram-negative bacteria that were on the fresh vegetable compete for predominance. *Enterobacteriaceae*, aerobic spore-formers, lactic acid bacteria, and other bacteria may be active. Eventually, the lactic acid bacteria gain predominance by lowering the pH, and primary lactic fermentation occurs. During primary fermentation, five species of lactic acid-producing bacteria are active, listed in approximate order of their occurrence: *Streptococcus* (*Enterococcus*) *faecalis*, *Leuconostoc mesenteroides*, *Pediococcus cerevisiae* (probably *P. pentosaceous* and/or *P. acidilactici*, according to recent classification[130]), *Lactobacillus brevis*, and *Lactobacillus plantarum*. Although all five species are active during fermentation of sauerkraut,[118] which contains relatively low concentrations of salt (ca. 2.25%), only the latter three species predominate in fermentation of cucumbers, which contain higher concentrations of salt (ca. 5% to 8%).[48] *Lactobacillus plantarum* characteristically terminates the lactic fermentation, apparently because of its greater acid tolerance.[121]

During fermentation of brined cucumbers, lactic acid bacteria may grow within the cucumber tissue as well as the brine.[22] Gas composition of the cucumbers at the time of brining greatly influences the ratio of bacterial growth in the cucumbers and the brine.[25] Yeasts were found not to grow within the cucumber tissue, presumably because of their larger size, which prevented their entry through stomata of the cucumber skin.

Green olives contain inhibitors of lactic acid bacteria,[55,57,87] which are thought to influence fermentation of Spanish-type green olives.[44,88] Yeasts are not inhibited and predominate in the fermentation when the olives are neither properly lye treated nor heat shocked before brining.[44]

Various species of fermentative yeasts also are active during primary fermentation. If fermentable sugars remain after primary fermentation, these sugars may give rise to secondary fermentation dominated essentially by yeasts. Fermentative yeasts grow as long as fermentable sugars are available; this may result in severe gaseous spoilage (bloater formation).[32,39,88] During post-fermentation, growth of oxidative yeasts, molds, and bacteria may occur on brine surfaces of open tanks that are not exposed to ultraviolet radiation or sunlight.[33, 112] Vegetable brining tanks are typically uncovered and are held outdoors to allow sunlight to reduce or prevent surface growth. Surface growth does not occur in fermented and anaerobically stored green olives.[140] Attempts have been made to develop a suitable anaerobic tank for the cucumber-brining industry.[63,66,85]

Lactic starter cultures have been used commercially on a limited basis in sauerkraut, olives, cucumbers, and other products.[54] *Pediococcus cerevisiae* and *L. plantarum* have been used in pure culture or controlled fermentations of cucumbers,[43,47] and olives.[44] Although starter cultures have been used on a limited commercial scale for fermenting cucumbers over the past 10 years, they are not widely used.

51.3 FLORA CHANGES IN SPOILAGE

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Production of CO_2 in the cover brine of fermenting cucumbers by various bacteria, including heterofermentative lactic acid bacteria and fermentative species of yeasts, is associated with bloater spoilage. Even homofermentative lactic acid bacteria such as *L. plantarum* and *P. cerevisiae* produce sufficient CO_2, when combined with CO_2 from cucumber tissue, to cause bloater formation in brined cucumbers.[56] The major source of CO_2 production by homofermentative lactic acid bacteria is decarboxylation of malic acid, a natural constituent of pickling cucumbers.[103] It has been demonstrated that cultures that do not degrade malic acid will ferment cucumbers with reduced bloater damage.[104] Procedures have been developed to produce and isolate non-malate-decarboxylating mutants of *L. plantarum*.[24,26] Purging fermenting cucumber brines with nitrogen has been shown to be effective in preventing bloater formation.[20,47,56,60] Purging is now widely used by the pickle industry. Air purging also is effective in preventing bloater formation,[20,60] but can result in cucumber softening due to mold growth,[21,60,75] reduced brine acidity due to yeast growth,[123] and off-colors and -flavors unless the purging regimen is care-