

# Chapter 7

## Lactic Acid Bacteria and Fermented Fruits and Vegetables



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### 7.1 Overview

China has a vast territory and abundant resources with an annual output of hundreds of millions of tons of fruits and vegetables. The per capita consumption and export volume are among the highest in the world. However, China's annual losses in fruit and vegetable products amount to tens of billions of yuan due to the lag of processing technology and industrialization and the lack of storage and processing methods, and the cost of losses is also at the top in the world. As common microbes in fermented food, lactic acid bacteria not only can improve the flavor and storage of the product but also have certain health functions. Therefore, it can not only facilitate the preservation but also increase the added value and meet the market demand for product diversity by applying lactic acid bacteria to the processing of fruit and vegetables.

Fermentation technology is one of the oldest food preservation technologies in the world, and fermented foods are widely distributed all over the world, especially in some underdeveloped countries. Fermented fruit may be the first fermented food in human history. Hunter gatherers often eat fresh fruit, but they will also eat rotten or fermented fruit when food is scarce. Repeated consumption of fermented fruit may prompt our ancestors to consciously improve the flavor of the fermented fruit. Studies have shown that fermented beverages appeared in Babylon 7000 years ago (Stanton 1998).

The production of traditional fermented foods is often considered as unsanitary or unsafe. Although it does occur at some point, but it is often overstated. Many fermented foods can inhibit the growth of spoilage bacteria due to lower moisture or higher acidity and salt concentration. So the fermented foods have a certain degree of security. In addition, fermentation is a very appropriate food processing and

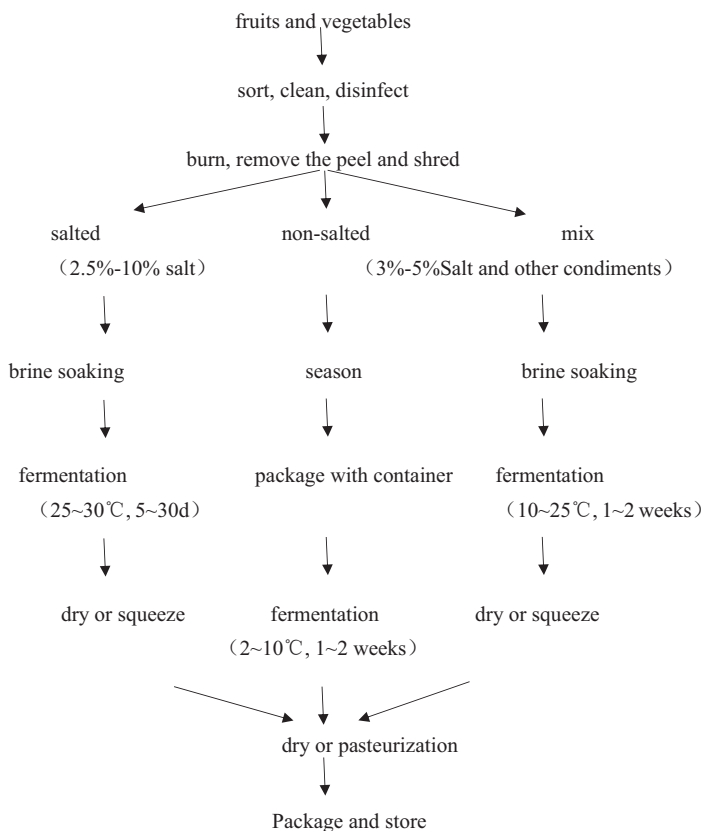
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preservation technology for developing or remote areas because fermentation technology is less dependent on refrigeration or other food preservation technologies. Therefore, natural foods or raw materials can be more effectively used, and waste materials can be even converted through microbial fermentation.

For the fermented foods like cheese, bread, beer, and wine, the production process is mostly in commercial scale, and the microbial fermentation processes had been studied in-depth. However, the relevant research and theory are still scarce for traditional fermented foods in Asia, Africa, and Latin America. Although the basic theory has broad consistency, the production conditions are quite different due to the diversity of fermentation products in different regions. Research on these production processes can be helpful to guide the product manufacture to improve the yield and quality of the fermented food.

Traditionally the fermented food is a natural fermentation process without sterilization. The microorganisms are selected in special environmental conditions and could produce unique end products. Lactic acid bacteria are the most widely used in fermented vegetables, and the fermentation process is shown in Fig. 7.1. Lactic acid



**Fig. 7.1** General process of fermentation of fruits and vegetables (Swain et al. 2014)

bacteria have long been used in vegetable fermentation, and China is one of the first countries to adopt this method for vegetable preservation. About 3000 years ago, Chinese people invented the production process of pickles, and the technology was transmitted to North Korea 1300 years ago which developed Korean kimchi that is popular in the world today. In Europe, lactic acid bacteria-fermented vegetables also have a long history. As early as the first century AD, people first reported the acid kimchi made from cabbage. In modern times, lactic acid bacteria-fermented vegetables are still very popular among the consumers at home and abroad due to its unique flavor and rich nutrition. Vegetables are rich in nutrients, including vitamins, cellulose, and minerals, but these ingredients are easily lost during the processing. However, the “cold processing” method of lactic acid bacteria fermentation is extremely beneficial to the maintenance of vegetable nutrients and color. At the same time, kimchi contains a large amount of lactic acid bacteria; after entering the digestive tract of the human body, it can relieve constipation by promoting gastrointestinal motility, lowering blood fat, and enhancing the host immunity. With the increasing in-depth research and development of lactic acid bacteria-fermented vegetables, traditional foods have become an emerging modern food industry.

Besides fermented vegetables, lactic acid bacteria-fermented fruit and vegetable juice is a new type of beverage developed in recent years. The research and development of lactic acid bacteria-fermented fruit and vegetable juice has broad prospects in enriching the beverage market and increasing the vegetable value due to the good flavor and nutritional health value.

## ***7.1.1 The Main Products of Fermented Fruit and Vegetables***

### **7.1.1.1 Fermented Vegetables**

Fermented vegetables are obtained under certain process conditions using beneficial microorganisms, including lactic acid bacteria, yeast, and acetobacteria, of which lactic acid bacteria are the most important microorganisms. Many vegetables can be used as raw materials for fermentation, such as cabbage, radish, kale, and green head. Common fermented vegetable products mainly include kimchi, sauerkraut, mustard, snow, fermented radish, and fermented olive oil.

### **7.1.1.2 Fermented Fruit and Vegetable Juice Drink**

#### **7.1.1.2.1 Fermented Fruit and Vegetable Juice Mix**

Among the current probiotic products on the market, fermented dairy products still occupy a dominant position, and 78% of probiotic products are produced and sold in the form of yogurt (Abu-Ghannam and Rajauria 2015). In recent years, vegetarian food has become more and more popular worldwide due to the high cholesterol

content in animal-derived products, and there are different proportions of patients with lactose intolerance in many countries. Nondairy probiotic products have become a new research hotspot. A variety of fruit and vegetable juices and cereal products are likely to be suitable carriers for lactic acid bacteria. Fruit and vegetable resources are extremely rich and inexpensive in China, but they depend on the seasons and can't be supplied all the year round. Fruit and vegetable juice are fermented by lactic acid bacteria and can be supplied all year round to meet the market needs. Moreover, the juices fermented by lactic acid bacteria such as *Lactobacillus plantarum* and *Lactobacillus acidophilus* are natural, nutritious, and healthy. Therefore, there are a large number of researches on the fruit and vegetable juices fermentation by lactic acid bacteria, including tomatoes, carrots, kale, pumpkins, and apples.

#### 7.1.1.2.2 Fermented Fruit and Vegetable Juice Milk Drink

Fresh and delicious fermented fruit and vegetable juice milk drinks can be produced by adding a small amount of milk powder and lactose. The strains used in the production process are the same as that used for producing yogurt, except that the amount of raw milk is reduced and 10–30% of the juice is added. The resulting beverage has both the frankincense of yogurt and the scent of fruits and vegetables and is more nutritious than yogurt.

### **7.1.2 Lactic Acid Bacteria Commonly Used in Fruits and Vegetables Fermentation**

#### **7.1.2.1 Homofermentative Lactic Acid Bacteria**

##### 7.1.2.1.1 *Lactobacillus plantarum*

The cells are nonmotile rods without flagella, occurring singly, in pairs or in short chains. Strains are usually facultative anaerobic and Gram positive. The optimum growth temperature range is 30–35 °C, and sugars are fermented to produce lactic acid. It plays a major role in the fermentation of fruits and vegetables due to its strong tolerance to acid and salt.

##### 7.1.2.1.2 *Streptococcus faecalis*

The cells are spherical, occurring in pairs or in chains. The strains are facultative anaerobic and Gram positive, and they do not produce spores. The growth temperature range is 25–45 °C, and the optimum temperature is 37 °C. The strains usually have poor tolerance to acid, and some strains are pathogens to humans and animals.

#### 7.1.2.1.3 *Pediococcus pentosaceus*

The cells are spherical occurring usually in pairs and not in chains. The strains are facultative anaerobic and Gram positive, and they don't produce spores. The optimum growth temperature range is 25–40 °C.

#### 7.1.2.1.4 *Lactobacillus casei*

The cells are rod without flagella. The strains are facultative anaerobic and Gram positive, and they don't produce spores. The optimum growth temperature is 37 °C, and glucose is fermented to produce acid. The strains have good tolerance to salt.

### 7.1.2.2 Heterofermentative Lactic Acid Bacteria

#### 7.1.2.2.1 *Leuconostoc mesenteroides*

The cells are spherical or elliptical occurring in pairs or in chains, usually in the form of short chains. The strains are facultative anaerobic and Gram positive, and they don't produce spores. It can grow at 10–32 °C, and the optimum growth temperature is 20–30 °C. It can ferment sucrose to produce a characteristic and viscous dextran capsule. It grows faster in the early stage of fermentation and plays an important role in the start-up stage of fruit and vegetable fermentation. The pH drops rapidly and inhibits the growth of spoilage bacteria. The acid, alcohol, and other substances produced during the fermentation could be combined with other metabolites and play an important role in the formation of flavor of the product. However, the strains have poor tolerance to the acid and the growth is slow in the later stage. *Leuconostoc mesogenes* can convert excess sugar into mannitol or dextran, while mannitol and dextran can only be used by lactic acid bacteria and cannot be used by other microorganisms or react with amino acids to form aldehyde or ketone groups. Therefore, it does not cause browning of food. *Leuconostoc mesenteroides* relies entirely on the pentose phosphate pathway to degrade glucose due to the lack of aldolase and isomerase.

#### 7.1.2.2.2 *Lactobacillus brevis*

The cells are straight rods with rounded ends occurring singly or in short chains. The strains are facultative anaerobic and Gram positive, and they don't produce spores. The growth temperature is 15–40 °C, and the optimum growth temperature is 30 °C. The strains can ferment pentose and form a unique flavor.

#### 7.1.2.2.3 *Lactobacillus fermentum*

The cells are usually short rods occurring in pairs or in chains. The strains are facultative anaerobic and Gram positive, and they don't produce spores. The growth temperature is 30–35 °C. It can grow above 45 °C and can't grow below 15 °C. The strains usually distribute on the surface of plants and in the gastrointestinal tract of animals and can survive in low pH and high bile acid intestinal environment. It has a promoting effect on regulating the balance of host intestinal flora and host health. The strains can ferment glucose to produce lactic acid, acetic acid, and ethanol, which promote the formation of flavor of the product (Zhang Yan-chao 2008).

### 7.1.3 *Nutritional Value and Functional Characteristics of Fermented Fruits and Vegetables*

#### 7.1.3.1 **Effects of Lactic Acid Bacteria on Nutritional and Physicochemical Properties of Fermented Fruits and Vegetables**

##### 7.1.3.1.1 Improve the Nutrition of Products

Lactic acid fermentation is a cold processing method, and the fermentation process does not reduce the nutritional value of vegetables. On the contrary, lactic acid bacteria can produce a variety of amino acids, vitamins, and enzymes by using soluble substances in raw materials. Lactic acid bacteria also have a weak decomposition ability for fat, which can increase the amount of free fatty acids in food. Wen-hui and Bin (1995) used *Lactobacillus bulgaricus* and *Streptococcus thermophilus* to ferment carrot juice and tomato juice and the content of vitamin B1, vitamin B2, and pyridoxine increased greatly after fermentation. Moreover, the content of glutamic acid and aspartic acid in the fermented product is increased, and the content of sulfur-containing amino acids like methionine and histidine is decreased. The calcium and phosphorus are in the form of ions after fermentation, which can be easily absorbed and promote the development of bones and can also prevent anemia and rickets caused by calcium deficiency and iron deficiency.

##### 7.1.3.1.2 Improve the Flavor of the Products

The fermentation products of lactic acid bacteria are mainly organic acids including lactic acid, acetic acid, and propionic acid, and these flavor substances can bring a soft sour taste. Ethyl acetate or ethyl lactate can be formed by combining lactic acid and acetic acid with an alcohol, imparting a fruity aroma to the product. Meanwhile, 2-heptanone and 2-nonanone can impart a certain fragrance and refreshing after

fermentation (Shin et al. 2003). In addition, the sour taste of lactic acid can mask the odor (green odor) of vegetable products (Ju-hua et al. 2003).

#### 7.1.3.1.3 Improve the Safety of Products

##### *Improve the Preservation of Products*

Fermentation is a low-cost, energy-saving food preservation technology. Fruit and vegetable products are prone to spoilage after harvest, especially in humid tropical regions, where special environments can accelerate the process of corruption. In order to prevent the corruption of food, there are drying, freezing, canning, pickling, and other processing methods. Among these methods, the freezing method has higher requirements on equipment, and the cost is also higher. The fermentation of fruits and vegetables does not require complicated equipment, and the low-acid environment after fermentation is also beneficial to product preservation. In addition, lactobacillin, hydrogen peroxide, and diacetyl formed during the fermentation have been proven to inhibit the growth of some spoilage bacteria and pathogenic bacteria. In recent years, lactobacillin has become a hot topic all over the world and is used as a natural preservative in the food industry. At present, nisin has been used as a natural preservative to extend the shelf life of foods, which has a broad-spectrum inhibitory effect on Gram-positive bacteria. Micrococccin can also inhibit the growth of Gram-positive bacteria, but it has not yet been approved by the food safety department. Thus both lactic acid bacteria and their metabolites can effectively prevent food spoilage (Martínez-Castellanos et al. 2011; Trias et al. 2008).

##### *Remove Anti-nutritional Factors or Toxins*

Some fruits and vegetables contain naturally occurring toxins and anti-nutritional compounds that can be detoxified or degraded by the fermentation of microorganisms. For example, cassava naturally contains cyanogenic glycosides, and it will release deadly cyanide in the body without proper treatment. After cassava is peeled and fermented for a certain period, toxic substances can be degraded by 90–95% (Battcock 1998).

Many fruits and vegetables generally contain a certain amount of nitrate, and some bacteria can convert nitrate to nitrite during the fermentation. Nitrite can be further converted to nitrosamine after intake, which is harmful to the human body. Lactic acid bacteria in fermented fruits and vegetables can reduce the formation of nitrite in fruit and vegetable products by inhibiting the proliferation of harmful bacteria and degrading nitrite. Lactic acid bacteria such as *Lactobacillus plantarum*, *Lactobacillus brevis*, *Lactobacillus fermentum*, and *Leuconostoc mesophila* were isolated from Chinese kimchi by Yan et al. (2008) and can degrade nitrite by more than 97%. Guang-yan et al. (2006) found that the content of nitrite in kimchi inoculated with *Lactobacillus plantarum* was significantly lower than that of naturally fermented kimchi.

### 7.1.3.2 Functional Properties of Fermented Fruits and Vegetables

#### 7.1.3.2.1 Relieve Constipation and Diarrhea

Constipation is caused by abnormal mechanical movement of the large intestine, mainly manifested by difficulty in defecation, rectal swelling, incomplete emptying, and prolonged emptying time. Diet is one of the common causes of constipation, and non-amyloid polysaccharides in the large intestine can restore the balance of intestinal flora and help relieve constipation. Kimchi contains a large amount of lactic acid bacteria and can relieve symptoms of constipation by producing short-chain fatty acids and regulating the intestinal flora. Kimchi is an essential dish in the homes of people in southwestern China, and kimchi water is a common prescription for treating chronic diarrhea in rural areas. Kimchi water is beneficial for spleen and dampness. Ping-chun et al. (2010) found that kimchi water had significantly beneficial effects in the treatment of diarrhea for children, with an effectiveness of 87%, which was better than Pfeiffer with an effectiveness of 68%.

#### 7.1.3.2.2 Lower Cholesterol

Cholesterol is closely related to the occurrence of diseases such as hyperlipidemia, diabetes, and coronary heart disease. Studies have shown that lactic acid bacteria can absorb cholesterol into their own cell membranes. The bile salt hydrolase produced by the lactic acid bacteria can decompose the bile salt and precipitate together with the lactic acid bacteria cells, thus lowering the blood cholesterol level (Li-na and Chang-qing 2007). The mechanism of lowering blood lipids by lactic acid bacteria may mainly include the following aspects. Some salts formed by organic acids such as acetate, propionate, and lactate can regulate the fat metabolism by lowering plasma total cholesterol (TC) and low-density lipoprotein (LDL) and increasing high-density lipoprotein (HDL). The enzymes produced by lactic acid bacteria can inhibit the synthesis of endogenous cholesterol. Some lactic acid bacteria can colonize in the intestinal mucosa, and its metabolism can reduce the absorption of cholesterol in the intestine, which may be related to the assimilation of cholesterol by lactic acid bacteria. Lactic acid bacteria can convert absorbed cholesterol into cholate and promote its excretion (Ling-yan et al. 2004; Yong 2003). Choi et al. (2013) studied the fasting blood glucose and serum cholesterol levels in two groups of volunteers with different intakes of kimchi per day. They found that eating kimchi every day can reduce the fasting blood glucose and serum cholesterol levels in both groups and improve the antioxidant capacity. Moreover, the effect was dose dependent and better effects were observed in the group of the volunteers with higher intake of kimchi (210 g/day) than that with lower intake of kimchi (15 g/day).



#### 7.1.3.2.3 Enhance Immunity

Xiao-ran et al. (2010) studied the effects of fermented fruit and vegetable juices on the immune functions in mice. Three kinds of fermented juices were prepared by mixing orange juice, pear juice, apple juice, and carrot juice with the ratio of 1:1:1:1 and then inoculated with *Bifidobacterium bifidum*, *Lactobacillus bulgaricus*, or *Streptococcus thermophiles*. The results showed that the three fermented juices significantly increased the thymus index and spleen index of the mice, indicating that they enhanced the body's immunity. Moreover, it can promote the production of serum hemolysin antibody in mice and enhance the degree of delayed-type hypersensitivity reaction and the phagocytic ability of macrophages, thereby improving the body fluid and cellular immune function of the body.

#### 7.1.3.2.4 Anticancer

In recent years, the anticancer effect of kimchi has attracted the attention of many scholars. Choi et al. (Choi and Park 1999) extracted the effective healthcare ingredients from fermented cabbage with methanol and then fed the extract to mice with gastric cancer. The extract was found to inhibit the growth of 60.7% of gastric cancer cells and prolong the life of the mice in the experimental group. Studies have also shown that the weight of tumors in mice with transplanted cancer cells was reduced by half and 40% of cancer cells are inhibited to metastasize after 3 weeks of feeding kimchi extract (Wen-bin et al. 2006).

#### 7.1.3.2.5 Lose Weight

Kimchi is a high-fiber, low-calorie food. Regular consumption of kimchi can stimulate the secretion of adrenal hormones, accelerate the burning of body fat, reduce the accumulation of fat in the blood and liver, and enhance the function of spleen immune cells. Korean scientists have shown that the mice fed kimchi had only 145–149 mg/g fat, while the fat content of mice without kimchi was 167–169 mg/g and the total fat content in the blood of mice fed kimchi was 170–200 mg/kg, while the total fat content in the blood of mice without kimchi was 246.1 mg/kg. Subcutaneous fat test also demonstrated that mice fed kimchi showed significant weight loss.

#### 7.1.3.2.6 Antiaging

Lactic acid bacteria and their metabolites have strong antioxidant activity because lactic acid bacteria can produce superoxide dismutase (SOD). SOD is a kind of enzyme containing metal elements, which can remove excess superoxide anion

radicals generated during metabolism in the body and improve immunity, delay aging, and reduce fatigue. In addition, lactic acid produced by lactic acid bacteria can inhibit the growth of spoilage bacteria and pathogenic bacteria in the intestinal tract. To a certain extent, the carcinogens and other toxic substances such as H<sub>2</sub>S and sputum matrix produced by these harmful bacteria are reduced, and the aging process of the body was slow down (Zhang and Cao 2005).

## **7.2 Fermented Vegetables by Lactic Acid Bacteria**

### **7.2.1 Main Types of Fermented Vegetable Products**

#### **7.2.1.1 Fermented Pickles by Lactic Acid Bacteria**

Fermented pickles are a kind of food with unique flavors by natural fermentation or artificial inoculation, which enables lactic acid bacteria to ferment the soluble nutrients in vegetables. During the fermentation, lactic acid bacteria can ferment the soluble components of plants and some saline extracts to produce flavor components such as lactic acid. In addition, the lactobacillin produced during the growth and metabolism can also play an antiseptic role.

#### **7.2.1.2 Fermented Sauerkraut by Lactic Acid Bacteria**

Sauerkraut is mainly distributed in the vast areas of northern China and is a kind of fermented vegetable formed by brewing fresh cabbage in an airtight container with brine for some period. Traditional sauerkraut is produced by natural fermentation, and lactic acid bacteria play an important role in the natural fermentation. Lactic acid produced during the fermentation can give the sauerkraut unique flavors. In the process of pickling, inoculation of mixed lactic acid bacteria can effectively improve the flavor and product quality of pickles.

#### **7.2.1.3 Fermented Olive by Lactic Acid Bacteria**

Olive is a subtropical woody oil fruit with fleshy stone fruit native to west Asia and later expanded to Mediterranean coastal countries. Olive is rich in oleic acid, linoleic acid, linolenic acid, and other unsaturated fatty acids, and they are mainly n-3 unsaturated fatty acids, the proportion of which is most suitable for human needs. It has certain medical and health functions for the digestive system, cardiovascular disease, bone, and nervous system development. The salted olive fermented by lactic acid bacteria was table olive, which was widely consumed in Europe (Gang 2007). According to the process method, there are two kinds of fermented olive including Greek-style and Spanish-style. Lactic acid bacteria fermentation can

change the pH and total acidity of the olive and improve the palatability of the product. Compared with naturally fermented olive, the bitter taste of fermented olive by lactic acid bacteria inoculation was decreased, and the aromatic taste was increased with increasing concentration of various flavor compounds (Sabatini et al. 2008). The results of isolation and culture of lactobacillus in fermented olive indicated that *Lactobacillus coryniformis* and *Lactobacillus pentosus* were identified as the main lactic acid bacteria during the fermentation of olive (Aponte et al. 2012).

#### 7.2.1.4 Fermented Pickled Mustard by Lactic Acid Bacteria

Pickled mustard was a well-known Chinese vegetable product, which was called “pickled mustard” because water was extracted from vegetables by pressing method during the processing (Zeng-san 2005). The pickled mustard was the main pickling vegetables, along with French pickles and German *Brassica oleracea*. Pickled mustard was fresh, crisp, delicious, and nutritious, with a good taste of sour and special salty taste. Compared with traditional pickled mustard, those fermented by lactic acid bacteria had higher yield of lactic acid, more flavor substances, stronger aroma and taste, and better sensory characteristics (Yun-bin et al. 2012).

#### 7.2.1.5 Fermented Potherb Mustard by Lactic Acid Bacteria

Potherb mustard is cultivated in south and north of China. Potherb mustard is very nutritious containing 7.3 mg of carotene, 0.35 mg of vitamin B1, 0.7 mg of vitamin B2, 4 mg of niacin, and 400 mg of ascorbic acid every 500 g fresh potherb mustard. In addition to rich nutrition, potherb mustard also has a unique delicious flavor. Therefore, whether fresh or pickled, potherb mustard attracts many dinners, especially pickled potherb mustard which has a long history of production. Fermentation of pickled potherb mustard by lactic acid bacteria is an effective biochemical preservation method. The main lactic acid bacteria during the fermentation of potherb mustard were *Streptococcus thermophilus*, *Lactobacillus bulgarian*, *Lactobacillus acidophilus*, and *Lactobacillus plantarum* (Gang 2007).

#### 7.2.1.6 Fermented Dried Turnip by Lactic Acid Bacteria

The traditional dried turnip is mainly fermented by lactic acid bacteria, which has a higher salinity in the curing environment, forming a hyperosmotic dehydration effect, thus inhibiting the growth of spoilage microorganisms. In the process of curing, a series of biochemical changes caused by the metabolism of beneficial microorganisms attached to the surface of radish strip can promote its fermentation and maturation, forming unique taste and nutrition. Compared with the natural fermentation, the dried turnip inoculated with lactic acid bacteria could accelerate the decline rate of pH in the fermentation process, shorten the fermentation period,

inhibit the growth of mixed bacteria, and improve the flavor of the product (Xiangyang et al. 2016). In addition, some researchers isolated one *Lactobacillus sakei* from the dried turnip, which showed good cholesterol-lowering functions (Changjian et al. 2011).

### 7.2.2 The Microecology of Fermented Vegetable Products

The production of traditional Chinese fermented pickles mainly relies on natural fermentation, which mainly relies on the microorganisms attached to the surface of vegetables. In the brine immersion environment, various microorganisms naturally attached to vegetables grow rapidly and use the dissolved carbohydrates and other components of raw materials for metabolic activities. During the fermentation, the species and quantity of microorganisms continue to change, and metabolites like lactic acid, acetic acid, and ethanol and a variety of volatile flavor substances are produced, so as to obtain mature pickles with good flavors. Lactic acid bacteria play a major role on flavor formation, while other microorganisms also exist including yeast and molds.

Tao et al. (2015) studied the changes in the amount of lactic acid bacteria, yeast, acetic acid bacteria, and molds in the fermentation of traditional pickles. The number of lactic acid bacteria increased rapidly in the first 2 days after the start of pickles fermentation and then remained stable until the end of fermentation. Yeast, acetic acid bacteria, and molds increased in the initial stage of fermentation and then decreased gradually until disappearing. As can be seen from the numbers, lactic acid bacteria are the dominant microorganisms in the fermentation of pickles and have good acid tolerance. The anaerobic environment is formed by the consumption of oxygen in the mid-fermentation vessel and the carbon dioxide production by the metabolism of heterofermentation. Acetic acid bacteria and molds are aerobic microorganisms, so their growth is inhibited. In addition, the decrease of pH by the production of acids can also inhibit the growth of yeast and molds. Further analysis of the dynamic change of lactic acid bacteria revealed that *Leuconostoc mesenteroides* subsp. *mesenteroides* started the fermentation, followed by *Enterococcus faecalis*, *Lactococcus lactis* subsp. *lactis*, and *Lactobacillus zoeae*, and finally *Lactobacillus plantarum* and *Lactobacillus casei* terminated the fermentation process (Tao et al. 2012).

Traditional analysis method revealed that *Leuconostoc mesenteroides* and *Lactobacillus plantarum* were the dominant bacteria during the fermentation of Korean pickles. However, more bacteria belonging to *Leuconostoc* and *Lactobacillus* in pickles were identified with the use of molecular identification techniques, such as *Leuconostoc citreum*, *Leuconostoc gasicomitatum*, *Leuconostoc gelidum*, *Lactobacillus sakei*, *Lactobacillus brevis*, and so on. However, some researchers analyzed the microbial composition of commercial pickles by non-culture method and found that *Weissella koreensis* was also the dominant bacteria in pickles (Kim

and Chun 2005). Lee et al. (2005) found that *Weissella confusa*, *Leuconostoc citreum*, *Lactobacillus sakei*, and *Lactobacillus curvatus* were the dominant bacteria in the fermentation of pickles using the molecular method. These results suggest that the fermentation of pickles involved a variety of lactic acid bacteria, including *Leuconostoc*, *Lactobacillus*, *Lactococcus*, *Pediococcus*, and *Weissella*.

Park et al. (2014) analyzed the composition of lactic acid bacteria of 13 Korean pickles (with a pH of 4.2–4.4) by pyrosequencing and found that *Weissella koreensis* accounted for the highest proportion (27.2%) in the lactic acid bacteria of pickles, followed by *Lactobacillus sakei* (14.7%), *Weissella cibaria* (8.7%), *Leuconostoc mesenteroides* (7.8%), *Lactobacillus gelidum* (6.3%), *Leuconostoc inhae* (1.2%), *Leuconostoc gasicomitatum* (1.2%), *Weissella confusa* (0.3%), and *Leuconostoc kimchii* (0.3%). At the genus level, *Weissella* had the highest proportion of lactic acid bacteria (44.4%), followed by *Lactobacillus* (38.1%) and *Leuconostoc* (17.3%). *Weissella*, especially *Weissella koreensis*, were the dominant bacteria in pickles.

Wei (2013) studied the microbial community of Sichuan pickles and industrial pickles by building 16S rRNA gene library and found that the microorganisms were mainly *Lactobacillus* and *Pediococcus*, accounting for 88.4% and 10.1%, respectively. The dominant bacteria were *Lactobacillus pentosus*, *Lactobacillus plantarum*, and *Pediococcus damnosus*, which accounted for 50.4%, 16.3%, and 10.1%, respectively. In addition, *Lactobacillus paralimentarius*, *Lactobacillus sunkii*, *Lactobacillus brevis*, *Lactobacillus kisonensis*, *Lactobacillus acetotolerans*, and *Lactobacillus namurensis* were also detected. The main microorganisms in Sichuan industrial pickles were *Halomonas*, *Lactobacillus*, and *Vibrio*, accounting for 13.5%, 32.4%, and 12.1%, respectively.

### 7.2.3 Lactic Acid Bacteria in Fermented Vegetable Products

#### 7.2.3.1 Species of Lactic Acid Bacteria Commonly Found in Fermented Vegetable Products

The common lactic acid bacteria in fermented vegetables mainly include *Lactobacillus*, *Lactococcus*, *Pediococcus*, *Leuconostoc*, and *Weissella*. The dominant lactic acid bacteria in pickles are mainly *Lactobacillus* and *Leuconostoc*, in which *Leuconostoc* dominates in the early stage of fermentation and *Lactobacillus* rapidly grow and replace its dominant position in the later stage of fermentation (Tao et al. 2012). Specifically, *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus brevis*, *Lactobacillus pentose*, *Lactobacillus curvatus*, and *Leuconostoc mesenteroides* were the common lactic acid bacteria in the fermentation of pickles (Xiao-lin et al. 2011; Yuan-feng and Li-gen 2007). The species and quantities of lactic acid bacteria in the fermentation have a crucial influence on the flavor and the qualities of the product.

### 7.2.3.2 Effects of Lactic Acid Fermentation on the Flavor Qualities of Fermented Vegetable Products

Generally, the volatile flavor components detected in fermented pickles with pure inoculation are less than that in traditional naturally fermented pickles. In the fermentation of pickles by pure inoculation, acid was rapidly produced with short fermentation period, and thus fewer flavor substances were accumulated. As a result, the taste and flavor were inferior to traditional naturally fermented pickles. Chun-yan et al. (2015) studied the differences of flavor substances in natural or inoculated fermentation of pickles and found that there was no significant difference in flavor substances, but the content of organic acids was different. However, Dan-ping et al. (2015) found that the main flavor components of pickles were different from each other and only nonaldehyde was the shared flavor component of all kinds of pickles. Bong et al. (2013) found that pickles fermented by a mixture of *Leuconostoc mesenteroides* and *Lactobacillus plantarum* had better sensory properties and higher free radical scavenging abilities than the naturally fermented pickles.

Zu-fang et al. (2008) made the pickled mustard by artificially inoculating *Lactobacillus plantarum* lact-8 and *Leuconostoc mesenteroides* lact-2 and optimized the fermentation process to obtain the optimal fermentation conditions. The conditions were shown as follows: salt addition 8%, fermentation temperature 25 °C, and inoculation amount 2% (the ratio was 1:1 for the two strains). The pickled mustard prepared under this condition had excellent physical, chemical, and sensory properties and was significantly superior to pickled mustard by natural fermentation in terms of nitrites, free amino acids, lactic acid bacteria content, acidity, pH, fermentation time, and sensory quality (Table 7.1).

**Table 7.1** Comparison of qualities between natural fermentation and artificially inoculated fermentation pickled mustard (Zu-fang et al. 2008)

Quality indicators	Optimization of fermentation	Natural fermentation
Fermentation time/day	15	17
Terminal pH	4.20	5.15
Terminal acidity/%	0.432	0.315
Lactic acid bacteria content/(cfu/ml)	$2.127 \times 10^7$	$3.733 \times 10^5$
Free amino acid/(mg/kg)	22.076	21.765
Nitrite/(mg/kg)	5.63	22.54
Sensory quality	Light yellow, strong aroma, crisp and tender, stable quality	Yellow, aromatic, crisp, and difficult to control

## ***7.2.4 Technologies and Characteristics of Fermented Vegetable Products by Lactic Acid Bacteria***

Fermented vegetables are prepared by placing washed vegetables in containers and adding ingredients, so that soluble ingredients in vegetables and some brine extracts can be used by lactic acid bacteria for fermentation. Brined vegetables usually contain high concentration of salt up to 12–15%, while pickles contain low concentration of salt. Fermenting vegetables with lactic acid bacteria will not reduce the nutritional value of vegetables but can improve their nutritional value. Lactic acid bacteria do not destroy plant cells and tissues, nor do they break down proteins because of their low activities of cellulolytic enzymes and proteases. The nutrients for lactic acid bacteria are mainly soluble substances of plants and some saline extracts. Lactic acid produced during the fermentation can not only act as a preservative but also improve product flavors.

### **7.2.4.1 Fermented Pickles by Lactic Acid Bacteria**

According to the raw materials and processing techniques, fermented pickles can be divided into Chinese pickles (Pao cai), Korean pickles, Japanese pickles, and Sauerkraut.

#### **7.2.4.1.1 Chinese Pickles**

##### *Material Selection*

The basic requirement of material selection is high content of solid, such as Chinese cabbage, cabbage, or radish, which can exceed 20% in some materials. Physical impact resistance is a direct advantage for materials of high content of solids. It should be noted that the nutrients of materials can meet the requirements of lactic acid bacteria.

Garlic, ginger, chili, onion, and cloves can be added during the pickle fermentation, which not only help to improve the flavors but also exert antibacterial effects. For example, the addition of clove can significantly prevent the growth of bacteria and delay the pH decrease and inhibit the decline of hardness of pickles during the storage. In addition, clove also contains a large amount of flavone and flavonoids with good antioxidant effects (Xiang-yang et al. 2015; Xue-ping et al. 2011).

##### *Crafting Process*

##### *Classification*

Check the cabbage for pests, yellow leaves, and rotting and inedible vegetables, and then the roots are repaired.

### *Segmentation*

Cabbage is usually segmented into two or four lengthwise or into small pieces. Cabbage and radish are usually cut into small pieces, while garlic, ginger, and chili are cut into shreds.

### *Add Salt and Accessories*

Generally, the amount of salt is 2.0–2.5%, and the amount of sugar is 2–3%.

### *Inoculation*

Fermented vegetables are traditionally inoculated with old pickle saline, while modern fermented vegetables are inoculated with pure cultures of lactic acid bacteria. Lactic acid bacteria at the end of the logarithmic phase are inoculated into the processed vegetables, which helps to better control the product quality and shorten fermentation period. The cultures are generally *Lactobacillus plantarum*, *Leuconostoc mesenteroides*, or mixed culture of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*.

### *Fermentation*

After inoculation and blending, the vegetables are divided into containers, which are pressed tightly and sealed tightly with water. The fermentation temperature is usually 20–25 °C, and the fermentation time is around 2 days to decrease the pH to 4.2.

### *Preservation*

The fermented vegetables are placed at 1–5 °C cold storage for preservation and sailed through the cold chain transportation.

#### 7.2.4.1.2 Japanese and Korean Pickles

Kimchi is the main food dish in Korea. Korean kimchi is spicy, refreshing, and delicious, with Chinese cabbage as the main raw material and red pepper as the main ingredients. Fish sauce, spices, mushrooms, seaweed sauce, and other ingredients are often added to increase the flavors. Some researchers also added mustard and Korean parasitic dendrobium extract to develop kimchi products with special healthcare effects (Fig. 7.2) (Park et al. 2014). Chinese cabbage are salted at 1–5 °C overnight and then mixed with chili paste for natural fermentation. After fermentation, the mature kimchi are packed for sale. Korean kimchi was mainly made by the hypertonic effect of salt and the fermentation by lactic acid bacteria, which not only keeps the vegetables fresh but also improves the nutritional value and healthcare functions (Lim et al. 2013; Yun-rong 2002).



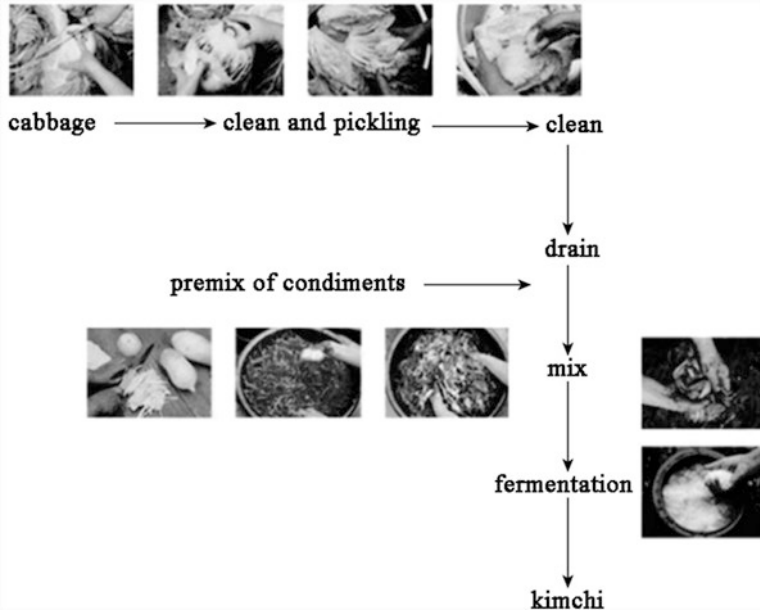


Fig. 7.2 Making of Korean fermented vegetables

Japanese kimchi is developed from Korean kimchi and has formed a unique style, which is different from Chinese kimchi and Korean kimchi in taste and production process. It was mainly made of natural pigments and soy sauce without the fermentation of lactic acid bacteria. Thus, Japanese kimchi belongs to a low-salt and low-acid and non-fermented product (Yan-gang and Quan 2011).

### 7.2.4.1.3 Sauerkraut

Sauerkraut is a traditional fermented cabbage in Germany and is now commonly found in Germany and Wisconsin State of America. The fermentation of Sauerkraut is a natural fermentation process dominated by the microorganisms on cabbage leaves. After fermentation, the plant tissue of cabbage is damaged, increasing the surface area and releasing nutrients for microbial growth under anaerobic conditions. The ion concentration and osmotic pressure are increased with the addition of salt in the juice, releasing nutrients from plant tissues and promoting the growth of required microorganisms. In the initial stage, the fermentation was mainly conducted by heterofermentative *Leuconostoc mesenteroides* and followed by the homofermentation of *Lactobacillus plantarum* in the late stage. During the fermentation, sucrose, fructose, glucose, and other carbon sources were transformed into lactic acid, acetic acid, ethanol, carbon dioxide, or mannitol, and the final fermented cabbage can be kept in a fermentation vessel for up to 1 year (Dijk et al. 2000).

### 7.2.4.2 Fermented Cucumber by Lactic Acid Bacteria

Cucumber is fresh and tender, fragrant, and delicious, containing protein, fat, sugar, cellulose, a variety of vitamins and calcium, phosphorus, iron, potassium, sodium, magnesium, and other rich mineral ingredients. In particular, cucumber contains special fine cellulose, which can reduce blood cholesterol and triglycerides, promote intestinal peristalsis, accelerate waste discharge, and improve human metabolism. Fermented cucumber is a popular fermented vegetable because of its crisp, sweet, and sour taste. The main process of its production was shown as follows: fresh cucumber → selection → sorting → cleaning → cutting → draining → bottling → adding salt water, white sugar, etc. → inoculation → brewing → bottling → seasoning → packaging → sterilization → finished products (Ying et al. 2015).

### 7.2.4.3 Fermented Sauerkraut by Lactic Acid Bacteria

Sauerkraut is a kind of fermented vegetable product with a long history. Sauerkraut is very fresh and delicious, increasing the appetite. The sauerkraut is rich in lactic acid bacteria with nutritional and health functions (Xiao-hui et al. 2009). The main process of its production was shown as follows: Chinese cabbage → drying and finishing → cleaning → entering barrel → crushing stone → filling water → fermentation → finished product.

### 7.2.4.4 Fermented Olive by Lactic Acid Bacteria

#### 7.2.4.4.1 Lactic Acid Bacteria in the Olive Fermentation

The lactic acid bacteria on the surface of olive include *Lactococcus lactis*, *Pediococcus pentosaceus*, *Leuconostoc mesenteroides*, *Lactobacillus plantarum*, *Lactobacillus delbrueckii*, and *Lactobacillus brevis*. In the past, natural fermentation was used by controlling the temperature, humidity, and concentration of salt solution. Now the fermentation is mainly inoculation by pure cultures. However, the olive fruit contains oleoresin (mainly glucoside), which inhibits the growth of lactic acid bacteria. So the variety characteristics of olive should also be considered during fermentation. Generally, *Lactobacillus plantarum* or micrococcus or mixed strains are mainly used for fermentation, and the effect of heterofermentation alone is not good (Gang, 2007).

#### 7.2.4.4.2 Process Flow

Olive → selection grading → water washing → removing → water washing → acid neutralization → water washing → heat → inoculation → fermentation → finished product

#### 7.2.4.4.3 Key Points of Olive Fermentation

A small amount of sugar can be added during the fermentation. Fermentation temperature should be kept at 20 °C with regular air sterilization. The fermentation can be ended while the pH arrived at 3.8–4.0 and the yield of lactic acid is above 0.5%. The fermented olive is yellow and moderate in hardness and has no bitter taste and salty taste.

#### 7.2.4.5 Fermented Pickled Mustard by Lactic Acid Bacteria

At present, high-salt pickling is commonly used in the production of pickle, and the batch salting process is mainly to prevent excessive salt solution from causing severe infiltration of pickling, resulting in sudden water loss and shrinkage of pickling tissue.

At the same time, vigorous fermentation can produce enough lactic acid, which inhibits the activities of other harmful microorganisms, which is not only conducive to the preservation of vitamins but also can shorten the time needed to reach the permeation balance and improve the curing effect (Yao-guang and Cheng-dong 1994). Natural fermentation adopts the method of high-salt pickling, while artificial fermentation adopts the method of low-salt pickling and inoculation with specific lactic acid bacteria starters. Some studies had shown that the artificial inoculation fermentation was significantly superior to the traditional natural fermentation, including reducing the use of salt, reducing the content of nitrite in products, improving the acidity of products, and inhibiting the contamination of miscellaneous bacteria (Shi-yang et al. 2013).

At present, the pickle pickling process is mainly divided into two kinds, namely, air dehydration and salt dehydration. Most of the pickle production in Sichuan and Chongqing adopts the way of wind dehydration, and the curing process is as follows: material selecting → stripping → pruning → wind dehydration → first curing → first pressing → second curing → second pressing → panning → mixing → loading and ripening. Most of the pickled vegetables in Zhejiang province are salted and dehydrated, and the curing process is as follows: selecting materials → stripping vegetables → cleaning → entering into the pond → adding salt → first application → turning the pond and adding salt → second pickling → ripening.

#### 7.2.4.6 Fermented Potherb Mustard by Lactic Acid Bacteria

The pickling process of potherb mustard is as follows: fresh cabbage → pretreating (selection, cleaning, etc.) → Pao cai → pickling → stepping vegetable → inverting cylinder or pond → sealing cylinder or pond.

Traditional potherb mustard preservation is a natural fermentation method; to join a high concentration of salt, most of the growth of microorganisms in the process of curing is restrained, but there is a small amount of microorganisms in growth;

some of the beneficial microbes has certain contribution to the flavor of potherb mustard; potherb mustard produced by the traditional curing process has good flavor. However, the fermentation cycle of traditional potherb mustard is long, and the operation process is complicated, which often makes the product quality difficult to reach the unified standard. For this reason, people adopted methods to improve the growth conditions of lactobacillus and add lactobacillus liquid and, combined with modern industrial production technology, greatly shortened the production cycle, simplified the production process, and increased the output (Wei 2005).

#### **7.2.4.7 Fermented Dried Turnip by Lactic Acid Bacteria**

Dried turnip is a traditional fermented vegetable and very popular in China, among which Xiaoshan dried turnip is the most famous. Its processing technology has a history of several thousand years, and it is the specialty of Xiaoshan district of Zhejiang province. Xiaoshan dried turnip is yellow and bright in color, crisp and refreshing, and nutritious and has anti-inflammatory, anti-heat, and appetizing effects. The processing method of dried radish in Xiaoshan is commonly known as air dehydration method, and the process could be summarized as follows: selection of raw materials → washing and cutting strips → white strips (fresh cut radish strips) → drying in the sun → salting and curing → loading altar with mixed materials (Zhi-qun 1986). At present, the production of dried turnip in China mainly adopts the traditional natural fermentation method with long fermentation period, high content of nitrite, and short shelf life. Therefore, the application of artificial inoculation of lactic acid bacteria has a broad prospect for dried turnip.

### **7.3 Fermented Fruit and Vegetable Juice Drinks by Lactic Acid Bacteria**

#### **7.3.1 Main Types of Fermented Fruit and Vegetable Juice**

##### **7.3.1.1 Fermented Fruit and Vegetable Juice**

Fermented fruit and vegetable juice is prepared by various fruit and vegetable juices as main raw materials and fermented by lactic acid bacteria or yeast alone or in combination. As a functional food, fermented fruit and vegetable juice by lactic acid bacteria has been studied in some developed countries including Japan, Korea, Germany, etc. It also has formed a great market development trend. The development of research on fermented fruit and vegetable juice is late and slow in China. There are few products in the market.

Fruit and vegetable juices are rich in nutrients, which will also increase after fermentation by lactic acid bacteria. Besides, the flavor in fermented fruit and vegetable juices will be improved. For example, carrot has rich nutrient because of carotene, vitamin B1, vitamin B2, and vitamin C. Tomatoes are nutritious and have a cool effect that promotes appetite. A variety of amino acids and polysaccharides are obtained in pumpkin, which can effectively prevent diabetes and high blood pressure. The germinated soybeans are rich in essential amino acids and minerals such as calcium, iron, zinc, and phosphorus. The health-promoting ingredients such as B vitamins and carotene are also multiplied. The fruit and vegetable pulp mixed by different fruit and vegetable juices is rich in nutrients and bright in color. After fermentation, its health function is greatly increased. So fermented fruit and vegetable juice is an ideal health drink. After acclimation, different strains of lactic acid bacteria can gradually adapt to the physical and chemical environment of compound vegetable pulp. They can use the nutrients in vegetable pulp to grow and produce flavor substances such as lactic acid. According to the needs of different groups of people, various fruits and vegetables are fermented separately, or several kinds of fruit and vegetable juices are combined to produce a variety of fermented fruit and vegetable juices, which can meet the demands and preferences of different consumers.

### **7.3.1.2 Fermented Fruit and Vegetable Juice Milk Drink**

The active lactic acid bacteria milk beverage is a new type of milk beverage that integrates nutrition and health functions developed in recent years. The product contains rich minerals, vitamins, proteins, and carbohydrates. The production process of fermented fruit and vegetable juice milk drink is similar to that of yogurt. Generally, *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* are used for fermentation. The amount of raw milk is reduced instead of 10–30% of vegetable juice. The beverages have lactic acid and milky aromas of yogurt, as well as the scent of vegetables, and are more nutritious than yogurt.

### **7.3.2 Microecology of Fermented Fruit and Vegetable Juice**

The microbial composition during fermentation is relatively simple because artificial inoculation is adopted by the fermentation of fruit and vegetable juice. Microbial community structure in natural fermentation enzymes was studied by Fang (Fang 2016), who believed that the microorganisms in the fermentation process of the enzyme mainly included bacteria and yeasts. The dominant bacteria mainly included *Lactobacillus harbinensis*, *Lactobacillus acetotolerans*, *Lactobacillus kefir*, *Acetobacter pasteurianus*, and *Bacteroides thetaiotaomicron*. *Pichia*, *Issatchenkia*, and *Saccharomyces* are the dominant yeast.

### 7.3.3 *Lactic Acid Bacteria in Fermented Fruit and Vegetable Juices*

#### 7.3.3.1 The Common Lactic Acid Bacteria in Fermented Fruit and Vegetable Juice

Recently, the mainly lactic acid bacteria used in fermented fruit and vegetable juice are *Lactobacillus* and *Bifidobacterium* including *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Lactobacillus acidophilus*, *Lactobacillus salivarius*, *Lactobacillus brevis*, *Bifidobacterium longum*, and *Bifidobacterium animalis* subsp. *lactis*. The common fruit and vegetable juice products fermented by lactic acid bacteria are shown in Table 7.2.

#### 7.3.3.2 Effects of Lactic Acid Bacteria Fermentation on Flavor Quality of Fermented Fruit and Vegetable Juice Beverage

The effects of lactic acid bacteria in the processing of fermented fruit and vegetable juices can be summarized as follows: improving the nutrients in vegetable juices, improving the flavor of vegetable juice, preventing the corruption of fruit and vegetable juices, extending their shelf life, and increasing the health benefits.

**Table 7.2** Examples of probiotics fermented fruit and vegetable juice

Material	Lactic acid bacteria	Reference
Tomato juice	<i>L. acidophilus</i> , <i>L. plantarum</i> , <i>L. casei</i> , <i>L. delbrueckii</i>	Yoon et al. (2004)
Carrot juice	<i>L. rhamnosus</i> , <i>L. bulgaricus</i>	Nazzaro et al.(2010)
Cabbage juice	<i>L. casei</i> , <i>L. delbrueckii</i> , <i>L. plantarum</i>	Yoon et al. (2006)
Beet juice	<i>L. acidophilus</i> , <i>L. casei</i> , <i>L. delbrueckii</i> , <i>L. plantarum</i> Y	Yoon et al. (2005)
Pumpkin juice	<i>L. plantarum</i> , <i>Saccharomyces cerevisiae</i>	Chun-li et al. (2014)
Ginger juice	<i>B. longum</i> , <i>L. casei</i> subsp. <i>casei</i> , <i>L. acidophilus</i>	Chen et al. (2009)
Litchi juice	<i>L. casei</i>	Zheng et al. (2014)
Apple juice	<i>L. paracasei</i> ssp. <i>paracasei</i>	Pimentel et al. (2015)
Coconut milk	<i>L. plantarum</i>	Pimentel et al. (2015)
Pineapple juice	<i>L. casei</i>	Costa et al. (2013)
Lemon juice	<i>L. casei</i> , <i>L. delbrueckii</i> , <i>L. plantarum</i> , <i>L. helveticus</i>	Islam et al. (2015)
Grenadine juice	<i>L. plantarum</i> , <i>L. delbrueckii</i>	Dogahe et al. (2015)
Banana juice	<i>L. plantarum</i> , <i>L. delbrueckii</i>	Tsen et al. (2009)
Cranberries, pineapples, orange juice	<i>L. salivarius</i> , <i>L. casei</i> , <i>L. rhamnosus</i> , <i>B. lactis</i> , <i>L. paracasei</i>	Sheehan et al. (2007)
Black raspberry juice	<i>L. brevis</i>	Kim et al. (2009)
Blackcurrant juice	<i>L. plantarum</i>	Luckow and Delahunty (2004)

For example, the flavor of tomato juice changed after lactic acid bacteria fermentation. The content of glutamic acid and aspartic acid was increased, which can increase the umami taste of the product. Some new flavor components were found by GC-MS after fermentation, which had a positive effect on increasing the fragrance and freshness of the product, including diacetyl, ethyl acetate, 2-heptanone, and 2-ketone. Low concentrations of diacetyl can present a creamy aroma, while esters of lower saturated fatty acids and fatty alcohols have a fruity aroma. Therefore, the flavor of fermented tomato juice is the combination of various components. The new components such as lactic acid, malonic acid, and succinic acid were found, and lactic acid was the highest organic acid in the fermented tomato juice (Li-hua and Jie-bin 1993). The relative content of carbonyl compounds in the volatile flavor components decreased by 61.6%, while the relative content of alcohol compounds increased by 2.4 times after the fermentation of pumpkin juice with lactic acid bacteria and *Saccharomyces cerevisiae*. The main flavor substances were ethanol, iso-amyl alcohol, and phenylethyl alcohol. The content of esters and organic acids in the pumpkin juice after fermentation was significantly increased. It demonstrated that fermentation process had a significant effect on pumpkin juice.

### **7.3.4 Process and Characteristics of Fermented Fruit and Vegetable Juice**

#### **7.3.4.1 Lactic Acid Bacteria-Fermented Fruit and Vegetable Juice**

Typically, a variety of fruit and vegetable juice can be fermented by lactic acid bacteria, such as apple juice, tomato juice, carrot juice, and a variety of other vegetable juice.

##### 7.3.4.1.1 Fermented Tomato Juice by Lactic Acid Bacteria

###### *Preparation of Tomato Juice*

Tomatoes with the maturity of more than 90% and dark red color are selected as raw material. After washing, they are blanched in hot water at 90–95 °C for 3 min, so that the outer skin is soft. The main effect is killing the microorganisms on the surface of the tomato, destroying the activity of the enzyme, and increasing the juice yield. Then tomatoes are put into the juicer for juice. Tomato juice is not appropriate for the growth of lactic acid bacteria because of its high acidity. Therefore, the acidity is adjusted with sodium carbonate before the colloid mill is homogenized, so that the pH is about 6.5, and 3–4% of sucrose is added.

###### *Preparation of Starters*

The species used for fermentation should be based on actual conditions. *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* or *Lactobacillus plantarum*, *Lactobacillus acidophilus*, and other lactic acid bacteria

may be used in mixed fermented vegetable juice. Generally, the starters are first activated 2–3 times, and then the compound vegetable juice and skim milk powder are used as a medium for inoculum enlargement. A seed starter of  $10^8$  cfu/ml is used as a working starter, and the inoculum amount is usually 3–4%.

### *Fermentation*

After the fermenter was thoroughly sterilized, the treated tomato juice was placed, sterilized at a temperature of 90–95 °C for 20 min, and cooled to 43 °C for inoculation. The fermentation temperature is 40–43 °C until the pH is reduced to 4.0–4.5, and the lactic acid concentration reaches 0.85–1.00%. The taste is best and the fermentation can be ended.

### *Termination of Fermentation*

The temperature in the fermenter is rapidly raised to 70 °C or higher to kill the lactic acid bacteria and then to fill and cool. Another choice is that the fermented tomato juices are rapidly filled in bottles after fermentation and stored at a temperature of 1–5 °C. The fermented tomato juice is red and turbid with tomato flavor and lactic acid bacteria fermentation flavor and sweet and sour taste. The pH is 4.0–4.5 and the lactic acid concentration is 0.85–1.0%. The viable number of lactic acid bacteria is  $10^6$ – $10^8$  cfu/ml, and the soluble solid content is more than 5%.

#### 7.3.4.1.2 Fermented Carrot Juice by Lactic Acid Bacteria

Carrot is one of the main vegetables all over the country. Its price is low and it is rich in nutrients, minerals, and vitamins. The fermented carrot juice not only maintains the nutrient composition of carrot but also has the characteristics of lactic acid fermentation, which can improve the flavor and taste of carrot juice and improve the intestinal functions and immunities.

The process is as follows: carrot → cleaning → slicing → squeezing → filtering → sterilization → inoculation → fermentation → post-cooking → mixing → packing → inspection → finished product (Rui and He 2002).

#### 7.3.4.1.3 Fermented Purple Potato Juice by Lactic Acid Bacteria

Purple sweet potato is rich in anthocyanins, dietary fiber, selenium, iodine, zinc, and other minerals. In addition to the ingredients and functions of ordinary sweet potato, it also has a variety of special physiological health functions and is an important raw material in the fields of food, medicine, and cosmetics. China has rich purple sweet potato resources, but at present the domestic purple sweet potatoes are mainly used for fresh food and processing purple sweet potato red pigment and purple sweet potato total powder. Although the nutrient content of purple potato is more



abundant, but its flavor is not as good as ordinary sweet potato, and the processing variety is single. Therefore, the purple potato food developed for the majority of consumers has a good market prospect. In order to retain the nutrients and bioactive substances in purple sweet potato as much as possible and increase its flavor, artificial inoculation of lactic acid bacteria is adopted for fermentation, and the production of purple sweet potato series of nutritious food, which is of great significance to improve the utilization rate of raw materials, enriches product varieties and enhances product added value. Xing-zhuang et al. (2013) prepared the fermented purple potato juices using *Lactobacillus plantarum*, *Lactobacillus* Reitman, and *Lactobacillus brevis* as starters. The process conditions were optimized by sensory evaluation and pH as indicators. The most ideal fermented purple potato juice could be obtained at the conditions of the inoculum of 2.0% with the ratio of three *Lactobacillus* 1:1:1, fermentation temperature 18–28 °C, and fermentation time 7–15 days.

#### 7.3.4.1.4 Fermented Strawberry Juice by Lactic Acid Bacteria

Strawberry is a kind of berry, which is not easy to store at room temperature and easy to rot. However, after fermentation by lactic acid bacteria, the fermented beverage of strawberry juice not only has the unique fragrance of fermented fruit juice but also has the fragrance of fruit. The product is sweet and sour and at the same time is beneficial for storage, which has considerable economic benefits. The technological process is as follows: strawberry juice → pH adjustment to 6.5 → heat sterilization (95 °C, 15min) → cooling, inoculation (*Lactobacillus acidophilus*, *Lactobacillus delbrueckii* subsp. *bulgarian*, *Streptococcus thermophiles*) → fermentation → blending → heat sterilization → canning → finished product (Chun-bao et al. 2001).

### 7.3.4.2 Fermented Mixed Fruit and Vegetable Juice Beverage by Lactic Acid Bacteria

#### 7.3.4.2.1 Purple Sweet Potato Milk Beverage by Lactic Acid Bacteria

He-sheng (He-sheng and Hai-ping 2015) developed purple sweet potato milk beverage with purple sweet potato and fresh milk as the main raw materials. The manufacturing process was as follows: fresh milk → adding purple potato, white sugar, and stabilizer → homogenization → sterilization → cooling → inoculation → aseptic filling → heat preservation fermentation → cold fermentation leave → finished product. Through orthogonal test, the optimal process conditions were obtained as fermentation time 6.5 h, citric acid addition 0.4%, white sugar addition 8%, and purple potato addition 20%. The product was uniform in texture and delicate in taste, sweet and sour, with purple potato and lactic acid fermented aroma and rich milky aroma.

#### 7.3.4.2.2 Pear Milk Beverage by Lactic Acid Bacteria

Xu-guang et al. (2015) developed a pear milk beverage; the manufacturing process was as follows: mixing raw materials (fermented milk, pear juice, stabilizer, sweetener) → adding citric acid to adjust acid → preheating (50–55 °C) → sterilization → canning → after cooking → finished product. The optimum parameters were 20% pear juice, 10% sugar, 0.15% citric acid, and 0.4% stabilizer.

#### 7.3.4.2.3 Longan Mixed Fruit and Vegetable Juice Drink by Lactic Acid Bacteria

An-shu et al. (2012, 2013) studied the preparation process of longan carrot, tomato, fruit, and vegetable juice milk beverage. *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* were used as starters. Longan juice, carrot juice, and tomato juice were mixed at 3.5:5:1.5; seed solution was inoculated to 4% and fermented at 37 °C for 24 h. The fermented drink was orange-red, had longan and lactic acid fermentation flavor, tasted sweet and sour, and was refreshing and soft. On this basis, the bactericidal beverage was developed. It was found that the sterilization conditions of longan fruit and vegetable juice mixed directly could affect the stability of the final drink. The best sterilization conditions were at 100 °C for 30 min.

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