Chapter 3

Japanese *Sake* Takes Her Rightful Place in the Ranks of International Wine?

It seems that Japanese *sake*, especially *ginjo-shu* (special-designation *sake*), is getting more popular in the world, but will this boom last long? At the same time, I will show the relationship between alcohol beverages and our health, and I will also introduce some Japanese *sake* culture in this chapter.

3.1 *Kagami biraki* has international cool

National universities of Japan have become semi-independent of the national government and they are required to maintain their own unique identity. My former employer, Osaka University, decided to create overseas office so that they could engage in cultural exchange. They established the San Francisco Education and Research Center as a base of operations in North America. After that they established centers at Groningen in Holland, Bangkok in Thailand and Shanghai in China. At the opening ceremony of the San Francisco research center, we had a *sake* production company contribute a wooden barrel of *sake* to us. Figure 3.1 shows the *kagami-biraki* or *-wari* ceremony. *Kagami biraki* (鏡開き) means opening the wooden cover of a *sake* barrel with wooden mallets as a blessed event. After that *sake* in the barrel is served to guests.

American people seem to like to see Japanese traditional festive ceremonies like *kagami biraki*, because it is rare to see people with Japanese-style jackets called "*happi*" while opening a barrel with mallets.

Sake is indispensable to Shinto ceremonies in Japan. When we have a Shinto ritual, we dedicate *sake* to the gods as offerings. After the ritual, we drink the *sake* (called *omiki*) (Fig. 3.2) served back by the shrine, and we have a chance to ask for the happiness of our families or a bountiful harvest.

Fig. 3.1 *Kagami-biraki at an opening ceremony of Osaka University SF-Education & Research Center at the Asian Art Museum, San Francisco.*

Kagami-biraki means opening the wooden cover of *sake* barrel with wooden mallets at a blessing event. After that *sake* in barrel is served to guests. The *sake* barrel was provided by Gekkeikan Sake Brewing Co. LTD, Sacrament, CA, USA.

Fig. 3.2 *Omiki (left) and San-san-kudo sets (right).*

Omiki is consecrated to Shinto shrine and usually served to people on New Year's Day. *San-san-kudo* sets are used at a wedding ceremony. (Photos quoted from ja.wikipedia.org)

Sake is also used for the *san-san-kudo* ceremony (Fig. 3.2) of a Shinto wedding, in which the bride takes first and next the groom and then bride again that take 3 sips of *sake* from 3 different cups, to make a total of 9 sips. This is to strengthen the bonds between husband and wife.

3.2 Quality water and rice makes quality *Sake*

Fig. 3.3 *Underground water used in sake brewing.*

Left, underground water steeped in mountains is used for *sake* brewing at Kirei Sake factory; right two photos, underground water and an old well used at Hakubotan Sake factory, Saijo, Higashi-Hiroshima, Japan. Accesses to these underground waters are also provided to citizens who appreciate natural mineral water.

Of all the alcoholic beverages in the world, I am going to talk about Japanese *sake*. Quality water is necessary for *sake* brewing, as well as any other alcoholic beverage. Nada, Fushimi, and Saijo are known as the top three *sake* brewing areas in Japan. In Nada, underground water with a high amount of minerals from the Rokko Mountains in Kobe has been used in *sake* brewing, while semi-soft underground water from the Ryuo Mountains, Higashi-Hiroshima, has been used for *sake* brewing in the Saijo area (Fig. 3.3). Unlike wine, Japanese *sake* involves complex production techniques. That is because the primary ingredient in *sake* is rice. As I wrote in the previous chapter, the wine made from grapes is fermented directly by yeast. Grapes contain as much sugar as glucose (grape sugar), and are therefore sweet. If we introduce yeast, it ferments the sugars in the grapes and

converts them into alcohol directly. However, rice contains little glucose, only starch. These starches are comprised of many glucose units and thus we have to degrade starch to glucose by microbial enzymes.

The rice used for brewing *sake* is special *sake* rice and it is different from everyday table rice (Fig. 3.4). The *sake* rice grain is larger than ordinary rice and it is easier to polish. In the center of the grain, there is a starch component called *shinpaku*. The *shinpaku* is filled with white starch grains (Fig. 3.5). By using rice containing less proteins, less lipids, and less umami substances, the taste of final brewed *sake* becomes clearer and superior. Therefore, the rice is polished to remove the outside of the *shinpaku*, that is, any material except starch is polished off. Newly polished rice is steamed and fermented in a multi-step process (Fig. 3.6). Most of local *sake* factories brew *sake* in the winter to control the appropriate temperature of *moromi*, as alcohol fermentation by *sake* yeast is a thermogenetic reaction.

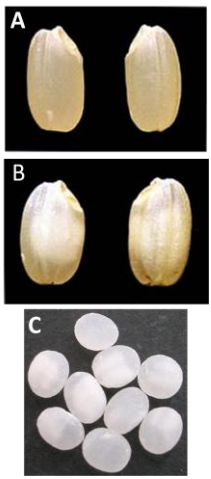

Fig. 3.4 *Sake rice grains.*

A, ordinary rice grains; B, *sake* rice grains; C, polished rice grains (shinpaku) from *sake* rice grains for brewing special *sake*, *ginjo-shu*. (Photos quoted from Yukawa-Syuzo Co, Kiso, Nagano, Japan).

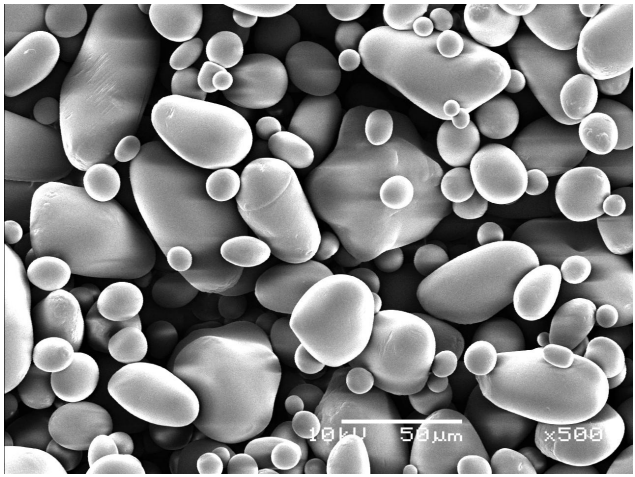

Fig. 3.5 *Electron microscopic picture of rice starch granules.*

(Picture provided by Dr. M. Hisamatsu, Mie University, Japan.)

Fig. 3.6 *Traditional and modern Sake brewing processes.*

Left and right panels show the traditional and modern *sake* brewing scenes, respectively. From top to bottom panels show rice steaming; *koji* preparation by mixing spores of *koji* mold and steamed rice; *shubo* preparation by mixing *sake* yeast with *koji* and steamed rice; and moromi fermentation sake making with *shubo*, *koji*, steamed rice and water are kept for 2 to 3 weeks at 10 to 12 ℃. The *sake* brewing process with modern technology is similar to the traditional methods. With modern technology, however, temperature, moisture and mixing conditions and time are controlled by computer and stainless steel tanks are used instead of wooden barrels. (Pictures provided by Dr. A. Nishimura, Hakutsuru-Syuzo Co. Ltd., Nada, Kobe, Japan).

3.3 *Ginjo-shu* is artistic *Sake* produced from extra-polished rice

There are two basic types of Japanese *sake*; ordinary *sake* and special-designation *sake*. The special-designation *sake* refers to premium *sake*s distinguished by the degree to which the rice has been polished. They are called *'Ginjo-shu'* or *'Dai ginjo-shu'* or *'Junmai-shu'* according to polishing ratio or ingredients. The rice polishing ratio for *'Daiginjo-shu'* is usually 50~60% (remains 50~40%) (Fig. 3.4). This special technique of *Ginjo-shu* brewery was first developed with soft water by Mr. Senzaburo Miura in Akitsu, Hiroshima. However, to polish rice too much seems to be a waste, isn't it? Therefore, I suggest that we should make special–designation *sake* out of unpolished rice by development of a new technique. If possible, it will be necessary for us to breed a new kind of rice variety, which contains only starch without the bran[*1].

The special-designation *sake*, *'Ginjo-shu'* in particular, is becoming more and more popular not only at Japanese-food-serving restaurants in New York or Paris but at various places in the world. It may be because Japanese *sake* is mild and has flavor of fruit. It doesn't have so much body as wine, so it is fit for French dishes which consist of oysters or fishery foods. The fruity flavor of '*Ginjo*- and *Dai ginjo-shu*' comes from fermentation with *koji* mold, *sake* yeast and lactic acid bacteria. It also comes from using of pure starch of *shinpaku* which doesn't have miscellaneous substances of rice. The alcohol percentage of ordinary Japanese *sake* is around 15-16%, but improved *sake* lowered to 12~14% seems to be liked by overseas people.

3.4 Traditional *Sake* brewing nurtured high skill of biotechnology

Sake yeast can convert glucose to alcohol but doesn't have the amylase which is necessary for degrading starch to glucose. So it must undergo a process of multiple fermentations. Here, a mold called *koji* appears which has many enzymes including amylase. I will write about it in Chapter 7. We call the process of conversion of starch to glucose 'saccharification'. This saccharification and alcohol fermentation by yeast in *moromi* mash occur simultaneously (Fig. 3.6).

During this process naturally habited lactic acid bacteria grow and produce lactic acid, which prevents the growth of miscellaneous contaminated microorganisms and help to work by pure yeast. *Kimoto* method of *sake* fermentation is known as the Japanese traditional orthodox method for preparing the starter mash. Natural lactic acid bacteria living in *sake* brewing houses or *sake* barrels have been used as a starter. Even now, some *sake* brewers are using this method in spite that it takes time, but most brewers add pure cultured lactic acid bacteria or lactic acid made by lactic acid fermentation. On the other hand *Lactobacillus fructivorons*, which makes *sake* sour as called *'hiochi'*, and by which *toji*, or brew masters were troubled for a long time, is another species of *Lactobacillus* though they are kinds of lactic acid bacteria.

After completion of the fermentation, the *moromi* mash is filtrated and sterilized to kill putrefying bacteria at 60 to 65 ℃ (140 to 149 ℉). The description of this sterilization method is seen as *hi-ire* technique on an ancient book, 'Tamon-in Nikki' described in AD 1478-1618. After that, about 300 years later, this sterilization technique was valued as a good pasteurization technique by Dr. Louis Pasteur in France.

Alcoholic beverages which are not distilled don't contain much alcohol compared with distilled beverages. For instance, the alcohol content of beer is about 5%, and in case of wine, around 13%, whereas in case of Japanese *sake*, about 15-16%. The alcohol content of Japanese *sake* can be heightened to 20% by the technique of *sake* production. This *sake* production process without distilling is, I think, No.1 in the world. The brewing process of *sake* production differs from the process of other beverages. For example, in case of beer, the conversion from starch to sugar and from sugar to alcohol occurs in two sequential steps. However, when *sake* is brewed, these conversions occur simultaneously. This technique has been kept with care and we can say that it is a result of traditional biotechnology.

3.5 Various kinds of alcoholic beverages in the world are different from Japanese *Sake*

Here, I would like to introduce various alcohol beverages which are different from Japanese *sake*. Beer, whiskey and vodka are made mainly from barley, wheat and rye, respectively, or mixtures of grains like corn, rice, *etc.* The main materials of those beverages are starch of barley and wheat, but malt, not *koji*, is used to convert polysaccharide of barley starch to glucose and maltose. Malt is germinated cereal grains that have been dried in a process known as "malting". Malt is the origin of maltose or malt-whiskey. Malt, like *koji*, has a lot of enzyme called amylase which degrades the grain's starches into sugars. Amylase contained in malt break down barley starch into glucose and maltose and the yeast converts these sugars to alcohol.

By the way, whiskey and vodka are distilled after fermentation. Alcohol is evaporated at lower temperature than water, so it can be concentrated by

distillation. This is what is called, a 'distilled' or 'spirit' beverage. Brandy is a distilled beverage of fruit liqueur like wine. Cognac is a kind of brandy produced at Cognac area in France. Although it is not a distilled beverage, champagne is a sparkling wine solubilizing carbon dioxide produced in Champagne District. Similar sparkling wine produced in other countries can't be allowed to call it 'Champagne'. Distilled beverages have high alcohol contents and some of them are higher than 40% alcohol. Japanese *shochu*, *awamori* of Okinawa and Korean *soju* are distilled alcoholic beverages and are popular in Japan and Korea. They are produced by using the ingredients of rice, barley, potatoes, *etc*.

Speaking of a spirit, when I visited in Poland I was offered a few kinds of good vodka at dinner. They seemed to be too strong for me and I was soon in paradise. I had the same experience when I drank *Baijiu* (白酒) at dinner time in China. *Baijiu* is produced from cereals, gaoliang (高粱), and its alcohol content is about 50%.

3.6 Unforgettable memory of margarita, tequila cocktail

Bourbon whiskey is a distilled beverage made from corn and it is produced in Kentucky State. Wild Turkey, Four Roses and General Grant are popular in Japan. Tequila is also a distilled beverage produced in Mexico. It is made as follows; first, inulin, which is a kind of polysaccharides and is contained in the pina of the blue agave plants (*Agave tequilana.* Fig. 3.7) like a cactus, is steamed and converted to fruit sugar and then fermented into alcohol by *Zymomonas* bacteria or yeast. Fermentation by non-yeast bacteria is rare and *Zymomonas* doesn't have much ability of alcohol fermentation. Therefore, it is distilled at least twice and the alcohol content is heightened.

Fig. 3.7 *Maguey as an ingredient of tequila production.*

Tequila is produced from pina (right) of *Agave tequilana* (left) fermented with *Zymomonas* bacteria. The fermented broth is at least 2 times distilled to concentrate alcohol. (Photos quoted from ja.wikipedia.org.)

Several years ago, we enjoyed our holidays at Cancun resort along the Caribbean Sea in Mexico. The view from there, emerald green seawater and tropical forests was marvelous and I drank tequila cocktail looking around the beautiful scenery. Since then, I became a fun of lovely-named delicious Margarita.

3.7 Biofuels were once Japanese specialties in the World War II

Although I digress from the main subject, I will write about biofuels here considering the problem of global warming. There is a movement that biofuels should be used as a replacement for fossil fuels. The reason why we call it 'bio-' is that ethanol or other biofuels are made by the technique of bio-technology, that is, fuels including ethanol can be synthesized chemically. In the USA, gasoline mixed with bio-alcohol is called 'gasohol' as a nickname. First, we make ethanol from starch of wheat, corn, sugar, fruits, *etc.* and use it as replacement fuels of gasoline. As a result, a problem was caused, that is, the

price of grains went up. Therefore, nowadays, cellulosic biomass such as bagasse or weeds came to be used as the method of production of ethanol. Biofuels have been successfully made of sugar cane in Brazil.

However, the technique of biofuels was originally Japanese specialties. When it became difficult for Japan to import oil from abroad before and during the Second World War, fuels for cars and aircrafts became scarce. At that time, scientists who were engaged in fermentation started the research of biofuels with yeasts or *Clostridium*. They tried to make a replacement for gasoline or diesel oil, such as ethanol, butanol and acetone from starch of potatoes or cellulosic biomass. In Great Britain, the famous Spitfire that defended the skies over Great Britain during World War II also used bio-butanol produced from potatoes. However, we cannot say for certain that the technique of fermentation of bio-alcohol or bio-diesel, which is drawing the attention of the world, was greatly improved compared with the technique of those days.

Whether biofuels are accepted or not from now on depends on the energy strategies of each country since biofuels do not become cheap and is not so much economical as the energy balance to produce them.

3.8 Is *Sake* the best medicine?

Drinkers have been insisting on the merits of alcoholic beverages saying, "Good wine makes good blood". I will tell you whether it is true or not from the view of a scientist. Alcoholic beverages contain various chemical compounds having physiological activity made of starch of grains fermented by *koji* mold (or malt), yeast and lactic acid bacteria.

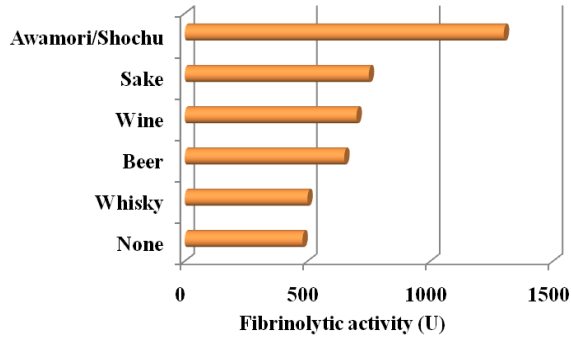

Fig. 3.8 *Effect of various alcohol drinks on thrombolytic activity.*

Men of 22 to 48 years old drank alcohol (30 to 60 ml) for 10 min and then determined their fibrinolytic activities one hour later. Numbers of examinees were 113 for non-alcohol drinker and for whisky, beer, wine, *sake*, and *awamori/honkaku-shochu* were 18, 41, 37, 37, and 62, respectively. (Report from Japan Sake Brewing Association, 2002. These data were based on the report by Dr. H. Sumi, 1988.)

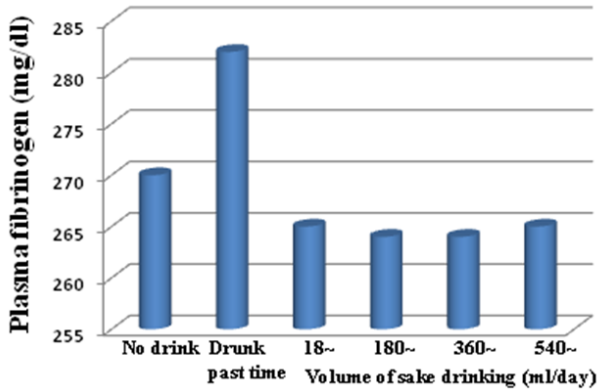

Fig. 3.9 *Effect of sake drinking volume on fibrinogen content in blood.*

Men of 40 to 59 years old drank designated volume of *sake* per day and determined fibrinogen in blood for 995 examinees. Data were compensated for age and smoking experience. (Report from Japan Sake Brewing Association, 2002. These data were based on the report by S. Sato et al., 1996.)

The merits of alcoholic beverages, which are worth of our notice, are that Japanese *sake* and aging *shochu* have a function as thrombolytic agent. Please

look at Figure 3.8. It shows the difference of the amounts of fibrinolytic enzyme in blood between persons who drank water without alcoholic beverages and persons who drank the same amount of each alcoholic beverage. Fibrinogen is a factor which forms *fibrin,* which is involved in the clotting of blood. It was surprising that Japanese *sake* and *awamor*i or *honkaku-shochu* contained the most amount of thrombolytic enzymes compared to other beverages. *Honkaku-shochu* is a distilled beverage without adding ethanol from outside and is aged for several years.

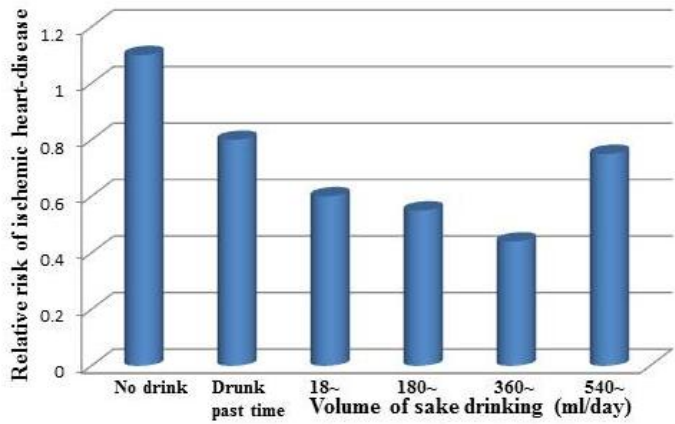

Fig. 3.10 *Effect of shochu drinking volume on ischemic heart disease.*

Men of 40 to 59 years old drank designated volume of *honkaku-shochu* per day from 1975 to 1984. Risk of ischemic heart disease was analyzed from the result of 8,476 examinees' drinking. (Report from Japan Sake Brewing Association, 2002. These data were based on the report by Dr. H. Ueshima et al., 1986.)

There is a data; if we drink Japanese *sake* (Fig. 3.9) or *honkaku-shochu* (Fig. 3.10) less than 2 *gou* (about 360 ml = 0.095 US gallons) per day, they lower the risk of cardiac myopathy. These data are very important in the point of analyzed living persons, but the data are rather old and no reconfirmed examination or the identification of effective materials has not been done since then. The volume of 360 ml *sake* or *shochu* seems too much for Japanese new

generations. However, an experiment was made on mice, and the result was that a little amount of alcoholic beverage is good for staving off aging. Of course, we should take care not to drink too much. Recently, lactic acid bacteria isolated from *kimoto*, the traditional seed mash used for brewing *sake*, showed anti-allergic effect[*2] and thus *sake* may have the ability to improve allergy symptoms mediated by the intestinal immune system.

3.9 Too much drinking causes various diseases

If scientists only emphasize the merits of alcoholic beverages, they should, at the same time, make people know that it has demerits which cause various diseases. There are various kinds of alcohol, but the only alcohol that we can drink is ethanol, which is consisted of 2 carbons. We must not drink one carbon methanol, 3 carbon propanol, 4 carbon butanol or others.

Alcoholic beverages have been said that they have adverse effects toward our health, like liver troubles, cancers, diabetes, gout, *etc*. The gastrointestinal surfaces are damaged if we drink when we are hungry. If we drink alcoholic beverages together with smoking, metabolic-syndrome is promoted furthermore. Alcohol is degraded into acetaldehyde by an enzyme in the liver and then turned into harmless acetic acid by acetaldehyde dehydrogenase. There are three kinds of this enzyme and one of them is lacking in the ability of degradation of acetaldehyde, which about 50% of the Mongoloids including Japanese people have, and as a result, in general, they are weak for alcoholic beverages. If we drink too much, acetaldehyde is stored in our liver and it causes hangover. The supplementary foods like sesamin, 5-aminolevulinic acid (ALA) or sulforaphane is said to be effective to hangovers by promoting the degradation of alcohol by activating the function of the liver and mitochondria. ALA is

multifunctional amino acid, which is an essential precursor of hem and vitamin B_{12} biosyntheses in our body[*3]. "Moderate drinking, by which our cheeks become slightly rosy and we can relax mentally and physically, is the best medicine" seems to be the truth.

3.10 Is it true that red wine prevents arteriosclerosis?

Probably you have often heard that red wine is good for health. There is an epidemiological investigation that French people have less risk of heart diseases like ring arteriosclerosis in spite that they take foods which contain much butterfat. This is called *'French paradox'*. It was thought that the reason was that they drink more red wine than people of other countries who take butterfat. Actually, it must be true.

Red wine contains various kinds of polyphenols more than white wine, because red wine is fermented together with grape peels. On the other hand, white wine is fermented without grape peels. Thus, red wine contains 6 times more polyphenols than white wine.

Arteriosclerosis refers to a thickening and hardening of arterial walls in the arteries. It is sometimes caused by the influences of smooth muscle tissues, that is, smooth muscles within the walls of blood vessels increases and moves. As a result, arterial walls get thickened and hardened. Catechin and epicatechin, which are main chemical substances of wine polyphenols, are said to inhibit the oxidation of LDL and to prevent smooth muscle tissues from moving about and also platelet-derived growth factor from binding their receptors. Therefore, we can say that red wine is good for health. However, you must not forget, 'Moderate drinking, about 180~360 ml (0.048~0.096 US gallons) a day, is important'.

3.11 Japanese *Sake* is being produced in the USA

By the way, how much Japanese *sake* do you guess is drunk in the world? The amount of Japanese *sake,* which is exported abroad, is increasing every year. The amount of export of Japanese *sake* is increasing in proportion to the number of increasing Japanese restaurants (Fig. 3.11). Export to Korea is increasing very much, in particular. A little diluted *ginjo-shu* will be drunk more in Europe and America. I hope it will be loved by a lot of people all over the world.

In the USA, Japanese *sake* brewing companies, such as Takara-shuzo, Ozeki-shuzo, Gekkeikan-shuzo, Yaegaki-shuzo and Hakushika-shuzo, are producing *sake* at several local places. These *sakes* are comparatively inexpensive prices by using rice grown in California. These *sake*s made in USA are also exported to Lain America and Europa. Figure 3.12 shows Japanese high school students in front of a *sake* brewing company located in Sacrament, California. They were invited to a tour of the biotechnology companies in the USA after they won the enterprise of bio-inter-high school sponsored by the Society for Biotechnology, Japan.

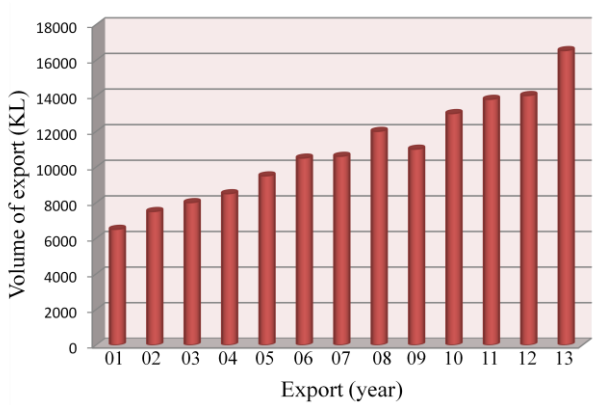

Fig. 3.11 *Export volume of Japanese sake.*

(Report from National Tax Administration Agency, 2012.)

Various labeled Japanese *sake* is being exported to the USA and some restaurants can serve hundreds of local *sake*. Generally, Japanese alcoholic beverages fermented from rice are generalized as *'sake'*, but I am afraid you cannot distinguish ordinary *sake* with special-designation *sake* such as *dai ginjo-shu, ginjo-shu, junmai-shu and josen-shu*. I hope you will ask a staff of the restaurant and enjoy delicious Japanese *sake*.

Fig. 3.12 *Visitors at a Japanese sake brewing company, Sacrament, CA, USA.*

3.12 *Ginjo-shu* should be protected by the international Commercial Law

Although the amount of exports of Japanese *sake* is increasing, it is one tenth of the amount of imports of wine to Japan. It is true that Japanese *sake* is minor yet, but I suppose it will be loved more all over the world, considering world-wide markets of alcoholic beverages are getting bigger. I think we have to advertise more positively that *ginjo-shu*, for example, is very tasty.

On the other hand, sham *ginjo-shu* may come into the market if exports of Japanese *sake* increase. As I touched it in the previous chapter, special-designation *sake* like *ginjo-shu* or *dai ginjo-shu* should be protected by

the international commercial law, like Champagne and Cognac are protected by AOC (Appellation d'Origine Contrôlée) in France. Special-designation *sake*, for example, *ginjo-shu*, must not be labeled so, if ratio of rice polishing is not observed and materials are not Japanese *sake* rice.

3.13 Summary

Japanese *sake* is produced from rice for *sake* brewing and clean water. The special-designation *sake* refers to premium *sake*s distinguished by the degree to which the rice has been polished. They are called *'Ginjo-shu', 'Dai ginjo-shu'* or *'Junmai-shu'*. It is fit for western dishes which consist of oysters or fishery foods. The process of conversion of starch in steamed rice to glucose by *koji* mold is necessary to produce *sake*. This saccharification and alcohol fermentation by yeast in *moromi* mash occur simultaneously. This traditional biotechnology has been kept with care and brought more than 16% alcohol accumulation without distillation. There are several reports that *sake* and aged *shochu* spirit lower the risk of cardiac myopathy. "Moderate drinking, by which our cheeks become slightly rosy and we can relax mentally and physically, is the best medicine" seems to be the truth.

[*1] Murooka, Y., Selective breeding of conventional and new industrial microorganisms: From Sake microorganisms to green algae. World Journal of Microbiology and Biotechnology, 8: 99-101 (1992).

[*2] Masuda, Y., Takayhashi T., Yoshida, K., Nishitani, Y., Mizuno, M., and Mizoguchi H., Anti-allergic effect of lactic acid bacteria isolated from seed mash used for brewing sake is not dependent on the total IgE levels. Journal of Bioscience and Bioengineering, 114:292-296 (2012).

[*3] Murooka, Y. and Tanaka, T., 5-Aminolevulinic acid (5-ALA): A multifunctional amino acid as a plant growth stimulator and stress tolerance factor. In: Anjun NA, Gill SS, Gill R (eds), Plant Adaptation to Environmental Change, p. 18-34, CAB International, Oxford shire, UK (2014).