

Fermentation and Sensory Characteristics of Korean Traditional Honey Wine from the *Saccaromyces sake*, *Saccharomyces bayanus* and *Nuruk*

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Saccaromyces sake, *Saccharomyces bayanus*와 누룩으로 제조된 전통벌꿀주의 발효 및 관능특성

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Abstract

This study investigated the characteristics of honey wine, which was fermented by *Saccaromyces sake*, *Saccharomyces bayanus* and *Nuruk*. For alcohol fermentation of the diluting solution, mixtures of *S. sake* and *Nuruk*, and *Nuruk* and *S. bayanus* resulted the excellent fermentation. These conditions also produced a higher alcohol content than that from the single yeast Strain. The values of pH and acidity of honey wine showed little changes during the fermentation processes. In the case of the mixture of *Nuruk* and yeast, the content of reducing sugar during the fermentation processes decreased rapidly, but the content of alcohol increased. The soluble solids were 7.5~8.1 °Brix after the fermentation period of 6 days, and the alcohol contents were represented as 12.5~13.1%, from the mixture of *Nuruk* and yeast. In addition, the sensory evaluation of honey wine with the mixture of *Nuruk* and yeast showed more effective results than that of the single yeast bacillus.

Key words : *Saccaromyces sake*, *Saccharomyces bayanus*, *Nuruk*, honey wine

Introduction

Honey wine well known as mead is a fermented drink produced by fermenting natural honey(1-3). Mead has a long history and its production was originated as far back as the ancient cultures of Egypt, Greek, and Rome approximately 2,000-2,500 years ago. It is directly fermented from the honey diluted with water. Its final alcohol content is about 12% and it had been usually used as a quality drink for a king or feudal lord in ancient times and the Middle Ages. In Korea, honey wine had been produced by fermentation with honey *Nuruk*(4). Although honey wine is widely produced in many

countries and had long history, it is not widely popular due to the lack of massivity, complication of fermentation process high level of sweetness caused by excessive unfermented sugar(5,6). Honey contains 0.34% protein, which causes decreased the quality of the final products due to increasing tubility in fermentation process and unpleasing burned or metallic smell in heating process(7,8).

Traditional mead was produced by diluting honey using only water, which are cause of improper acidity, bad fermentation, delayed the fermentation, and a lowered quality because honey do not have enough vitamin, inorganic and nitrogenous nutrients for the growth and fermentation of yeast(5).

In order to solve these problems, some fruit juices were added to the honey(6), and it was treated at a high temperature

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for a short time(9). In addition, the microfiltration method(10), developed to remove the protein in honey wine production. Several studies on the fermentation conditions of honey wine have been reported(11,12). Some fruit wines that are produced by adding honey to apples(13), grapes(14) and osmotic extracts(15,16) have been also investigated.

On the other hand, Korea has traditionally had an excellent brewing technology, and supplies drinking culture, such that it is extremely important that these traditional wines will be recovered, preserved, and cherished. Recently, the interest in traditional wines having high quality has been increased, since the standard of life and income levels were increased.

Therefore, honey wine, a Korean traditional wine, may become an important item to activate domestic bee farming industry. In this study, honey wine was produced according to the Korea's original production method as a basic study for the development of traditional home brews and their industrialization. This study also dealt with changes of the ingredients of honey wine in the production and fermentation processes by adding *Nuruk* and yeast mixtures to improve its quality.

Materials and Methods

Materials

Honey used containing a $19.0 \pm 1\%$ of moisture, $0.3 \pm 0.1\%$ of ash, $70.0 \pm 2\%$ of reducing sugar, 78 ± 2 °Brix of soluble solid, and 3.7 ± 0.2 of pH was kindly provided by GABO Co. Korea. *Nuruk* was purchased from SONGHAK Co. Korea. Two kinds of *Saccharomyces sake* (Japan Fermentation Associate) and *Saccharomyces bayanus* IFO 1802, were purchased from Difco, USA for the fermentation of honey wine.

Preparation of seed mash

The nutrients of yeast were reconfigured according to the references of nutrients of yeast that are presented by Rhim et al.(15) as following : to a 100 mL of honey liquid controlled at 10 °Brix were added 1.00 g of $(\text{NH}_4)_2\text{SO}_4$; 0.5 g of K_3PO_4 ; 0.20 g of MgCl_2 ; 0.05 g of NaHSO_4 ; 0.02 g of peptone ; 5.00 mg of thiamine ; 2.50 mg of Ca-pantothenate ; 2.00 mg of inositol ; 0.25 mg of pyridoxine ; 0.02 mg of biotin, and treated with a heat sterilization for 10 min under the pressure of 1.2 kg/cm^2 . Then, a 1 mL of yeast culture was inoculated in the liquid and cultivated at 25°C for 48 hr to cultivate a seed mash.

Preparation of honey wine

To a honey solution diluted by water to 24 °Brix of soluble solid content was added a 100 ppm of Na_2SO_3 used as a raw fermentation material after being left it for about 1 day.

Fermentation and filtration

The fermentation was initiated by adding 5% of seed mash, into the 8 L of the fermentation material controlled by 24 °Brix. In the first stage of fermentation, the mouth of the fermenter was covered by gauze to keep an aerobic state and kept at a temperature of 30°C. The solution was shaken sporadically and allowed to alcohol fermentation at 20°C in the anaerobic state by closing the fermenter after the bacillus had fully grown and then fermented for 2 days. *Nuruk* was fermented by adding an amount of 10% of fermentation material, controlled by 24 °Brix. The fermented honey wine was sterilized and clarified by a microfiltration method, using membrane filter (0.45 μm of poresize, DDS Mini-Lab 10, De Danske Sukkerfabrikker, DDS RO-Division, Denmark).

Physicochemical constituent analysis

The values of pH processed in the fermentation were measured by using a pH meter (Orion Research Inc. 520A, USA). The total acid was presented as the amount of citric acid presented by titrating at pH 8.4 with a 0.1N NaOH solution. The soluble solid was measured by units of °Brix using a refractometer (Atago Hand Refractometer, Japan). The reducing sugar was measured by the Somogyi method(17). The 80 mL of vapour solution obtained from 100 mL of each sample was adjusted to 100 mL of test solution with diluted water and its alcohol content was measured at 15°C(15).

Hunter value and transmittance

The Hunter value, consisted with L (brightness), a (redness), and b (yellowness) value was measured using the Hunter Lab Colorimeter (Color Quest Hunter Lab Associates Laboratory, Inc. USA). In order to examine the clarifying degree of each sample, the transmittance of the sample was measured at 660 nm a UV-Vis spectrophotometer (Hewlett Packard, HP 8452, USA).

Sensory and statistical analysis

Twenty panelists whose ages ranged from 20 to 50 years evaluated the sensory of each honey wine and scored their liking using seven-point hedonic scale(1, dislike very much, 2, dislike moderately, 3, dislike slightly, 4, neither like nor

dislike, 5, like slightly, 6, like moderately, 7, like very much). Each parameter was tested in triplicate. Statistical analysis were carried out using a SAS(Statistical analysis system for WindowTM). Treatment means were compared using Duncan's multiple rang test in SAS.

Results and Discussion

Changes of physicochemical constituent in the fermentation

Honey wine was fermented by adding *Nuruk* and yeast, *S. sake*, or *S. bayanus*, either as singular units, or as mixtures. Fig. 1 to Fig. 3 present the changes in the fermentation process.

The changes in the reducing sugar content was decreased steadily from the beginning of fermentation to the 18th day, as in Fig. 1. In the case of the single addition of *S. sake* (SS), the sugar was decreased by 50% after a period of 6 days, compared to the initial stage of fermentation, and decreased up to 70% in the latter period of fermentation. On the other hand, the content was reduced to 90% comparing with it of the initial stage of fermentation in the case of the single addition of *S. bayanus* (SB), the mixture addition of *Nuruk* and *S. sake* (NSS), and the mixture addition of *Nuruk* and *S. bayanus* (NSB). After 6 days of fermentation, the reducing sugar of each additional group consumed about 50% of the initial stage of fermentation for the SS, 55% for the SB, 69% for the NSS, and 78% for the NSB. Rhim et al.(12) reported that honey wine produced from honey only using of *S. uvarum* showed to reduce the 50% of the sugar after a period of 16 days and the plum melomel, fermented by honey and plum extract consumed about 80% of the initial reducing sugar after a period of 16 days.

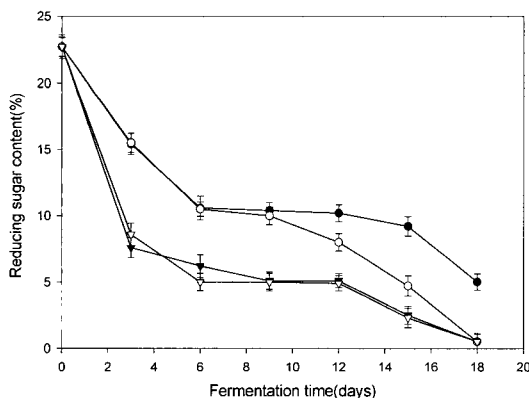


Fig. 1. Changes of reducing sugar content during fermentation.

Honey wine were prepared with yeast strains and *nuruk*-yeast mixture. ●-●, *Sacch. sake*; ○-○, *Sacch. bayanus*; ▼-▼, *nuruk-Sacch. sake* mixture; ▽-▽, *nuruk-Sacch. bayanus* mixture.

The reducing sugar on the cases of the NSS and NSB after a period of 6 days from the fermentation examined in this study were consumed to 70~80%, comparing with the values of the initial reducing sugar contents. From this result, it seemed that the speed of the reducing sugar for honey wine fermented by adding the mixture of *Nuruk* and yeast was about 10 days faster than that of the reported fermentation conditions(15,16). The reason of decreasing speed on the reducing sugar in the mixture of *Nuruk* and yeast, comparing with that of the single addition of yeast bacillus, may due to the adaptation of the strains in the initial stage of fermentation. Moreover, it also showed that are more active in the decreased speed of the reducing sugar and the amount of alcohol production, the fermentation of the mixtures of *Nuruk* and yeast, than that of the single addition of yeast, until the latter period of fermentation.

The changes of the soluble solid tended to decrease smoothly throughout the fermentation processes as shown in Fig. 2. The decreasing properties were similar to those of reducing sugar. In the case of adding a mixture *Nuruk* and yeast after 6 days of the fermentation, the contents of the soluble solid were 7.5~8.1 °Brix, which was lower than that of 14.5~16.4 °Brix in the single use of yeast.

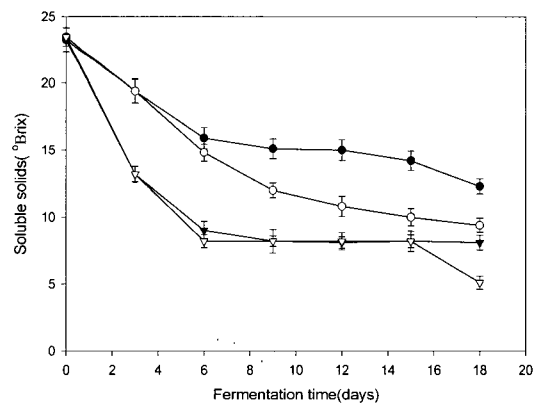


Fig. 2. Changes of soluble solids content during fermentation.

Honey wine were prepared with yeast strains and *nuruk*-yeast mixture. ●-●, *Sacch. sake*; ○-○, *Sacch. bayanus*; ▼-▼, *nuruk-Sacch. sake* mixture; ▽-▽, *nuruk-Sacch. bayanus* mixture.

Alcohol is produced by consuming the sugar contained in the fermentation solution. As shown in Fig. 3, the content of alcohol in the fermentation process presented various aspects, such as a smooth or rapid increase, or other different changes, between the single addition of bacillus, or the addition of the mixture of *Nuruk* and yeast. The alcohol contents in the latter period of fermentation, and after 18

days, showed 9.0% for the SS, 12.5% for the SB, 14.0% for the NSS, and 14.5% for the NSB. On the other hand, the amount of alcohol production in the single addition of yeast bacillus showed value between 5.2~7.2% in the case of a 6 day period of the fermentation.

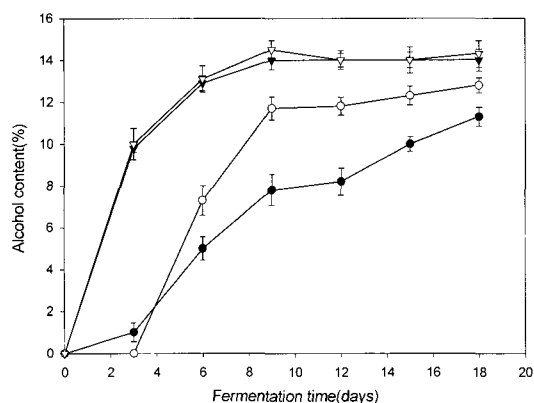


Fig. 3. Changes of alcohol content during fermentation.

Honey wine were prepared with yeast strains and *nuruk*-yeast mixture. ●-●, *Sacch. sake* ○-○, *Sacch. bayanus* ▼-▼, *nuruk*-*Sacch. sake* mixture ▽-▽, *nuruk*-*Sacch. bayanus* mixture.

While, the alcohol content in the addition of the mixture of *Nuruk* and yeast was 12.5~13.1%. In the honey wine produced with the mixture of *Nuruk* and yeast, the alcohol contents showed to 12.5~13.1% which were similar to the alcohol content of a regular wine. However, in the case of the honey wine produced by yeast only, a relatively low alcohol content and high sugar content were remained. This suggests that the fermentation was not thoroughly performed. From the results on the decrease of the reducing sugar in the case of a single use of yeast as shown in Fig. 1 and the tendency of the production of alcohol as presented in Fig. 3, it seemed that the fermentation continued to proceed was delayed in the middle of the fermentation. In addition, it was known that long term fermentation will affect the quality of the final products because it increases the possibility of contamination(3). However, this study showed that the optimal fermentation period of honey wine was reduced 6 days, from 10 days of the reported fermentation period(3,15) by adding the mixture of *Nuruk* and yeast. In the case of the single addition of yeast, the alcohol production presented was relatively small after a period of 6 days, but increased continually after that period. This is expected due to the tendency toward decreased the reducing sugar as shown in Fig. 1. It was regarded that the nutrients, which were needed to grow the yeast except for sugar, or enzymes for glycosidase

were not enough in the initial stage of fermentation. In the case of the addition of the mixture of *Nuruk* and yeast, the enzymes existed in the *Nuruk* acted as an accelerant to resolve the unfermented sugar.

Characteristics of honey wine

This study produced honey wine by adding a single yeast, or a mixture of *Nuruk* and yeast and examined the characteristics of the honey wine, after fermentation and a microfiltration in each fermentation condition (23 days for the fermentation of the SS, 20 days for the NSS, and 6 day for the NSS and NSB). Table 1 presents that the value of pH(3.37~3.62) and the total acids(0.40~0.42%) was not significant different. From the measurements of the colors of each additional group by using a spectrophotometer, there were little differences between the single addition of yeast and the addition of the mixture of *Nuruk* and yeast. It showed that both conditions were able to produce equal products. Applying a microfiltration method for the produced honey wine, the transmittances of the honey wine, which was applied by the single addition of yeast and the addition of the mixture of *Nuruk* and yeast, were 99.3~99.5%. Then, it was possible to produce a pure and transparent product. In addition, the sensory evaluation of honey wine with an added mixture of *Nuruk* and yeast showed more effective results than that of the single addition of yeast bacillus(Table 2).

Table 1. Characteristics of experimental honey wine

	SS	SB	NSS	NSB
pH	3.37	3.47	3.53	3.62
Total acidity(%)	0.41	0.40	0.40	0.42
Hunter Value L	46.23	45.53	44.34	44.91
a	0.30	0.34	0.28	0.34
b	5.98	5.97	5.98	5.63
Transmittance(%)	99.40	99.30	99.40	99.50

Honey wine were prepared with *Sacch. sake*(SS), *Sacch. bayanus*(SB), *Nuruk*-*Sacch. sake* mixture(NSS) and *Nuruk*-*Sacch. bayanus* mixture(NSB) and fermented during 23, 20, 6 and 6 days, respectively.

Table 2. Sensory quality¹⁾ of experimental honey wine

	SS	SB	NSS	NSB
Flavor	4.30 ^{a2)}	4.60 ^d	5.40 ^b	5.55 ^c
Taste	3.40 ^b	4.12 ^a	5.12 ^c	5.17 ^c
Color	3.23 ^a	4.32 ^b	4.99 ^c	4.87 ^c
Overall	3.64 ^d	4.35 ^b	5.17 ^c	5.20 ^c

Honey wine were prepared with *Sacch. sake*(SS), *Sacch. bayanus*(SB), *Nuruk*-*Sacch. sake* mixture(NSS) and *Nuruk*-*Sacch. bayanus* mixture(NSB) and fermented during 23, 20, 6 and 6 days, respectively.

¹⁾1: dislike very much, 7: like very much.

²⁾Mean separation in columns by Duncan's multiple range test at 5 level.

These results showed that there were differences in quality between the honey wines, which were fermented by the single addition of yeast bacillus and by the addition of the mixture of *Nuruk* and yeast.

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요 약

효모 *Sacch. sake*, *Sacch. bayanus* 그리고 누룩을 이용하여 벌꿀주를 제조하고 그 발효특성 및 관능적 기호도를 조사하였다. 벌꿀 희석액의 알코올발효에는 *Sacch. sake*와 누룩의 혼합첨가 그리고 *Sacch. bayanus*와 누룩의 혼합첨가에 의한 벌꿀 발효주가 효모 단독균주를 이용하는 것보다 발효력이 우수하게 나타났으며, 알코올 생성량도 높게 나타났다. 효모와 누룩 혼합첨가구의 경우 발효중 환원당은 급속히 감소하고 알코올 함량은 증가하여 발효 6일에 가용성 고형분은 7.5~8.1 °Brix이었다. 발효 6일에 알코올 생성은 효모와 누룩 혼합첨가를 사용한 경우가 12.5~13.1%로 단독 효모균주를 사용한 것의 5.2~7.2% 보다 높게 나타났으며, 발효도 단독 효모균주 첨가에 비하여 양호하여 단독 효모보다는 누룩과 효모를 혼합하여 첨가한 것이 1.8~2.4 배 더 빠르게 진행되었다. 벌꿀주의 관능적 기호도는 효모 단독보다는 효모-누룩 혼합첨가구에서 상대적으로 높은 점수를 얻었다.

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