MICROBIAL AND PHYSIOCHEMICAL CHANGES DURING THE INCUBATION OF *FEN-DAQU*

Le Van Diep[1, *], Han Bei-Zhong[2]

[1]*Faculty of Chemistry, Vinh University, 182, Le Duan Street, Vinh City, Nghe An province*

[2]*College of Food Science and Nutritional Engineering, China Agricultural University, China*

[*]Email: levandiep@vinhuni.edu.vn

Received: 19 October 2012; Accepted for publication: 24 February 2014

ABSTRACT

Fen liquor is typical of Chinese light-flavor liquor, which is fermented from sorghum with *Fen-Daqu* powder. *Fen-Daqu* is a saccharifying agent and fermentation starter obtained by natural solid-state fermentation under non-sterile conditions. The standard plate count and the online measurement methods were used to enumerate the surviving microorganisms, and measure the physiochemical in *Fen-Daqu* during the incubation. Total counts of mesophilic aerobic bacteria (30 °C and 55 °C), bacterial endospore (30 °C and 55 °C), lactic acid bacteria, enterobacteriaceae and fungi starting with minimum level around 10^6, $<10^4$, $<10^5$, $<10^5$, 10^5, $< 10^4$ and 10^5 cfu/g and attaining maximum around 10^{11}, 10^9, 10^9, 10^9, 10^7, 10^5 and 10^8 cfu/g, respectively. During the incubation of *Daqu* the microorganisms increased from *Woqu* to *Liangmei* periods and gradually decreased during the later phases. The pH in *Daqu* was increased over time during the incubation. The total acidity in *Daqu* increased and reaches to maximum at *Shangmei* phase (around 4.5 g lactic acid per kg *Daqu*) and then gradually decreased over the time. The relative humidity in incubation room was reduced from around 100 % to around 20 %. Temperature in incubation room was increased over time from the first to middle period and decreased in the *Yangqu* phase. Temperature in *Daqu* inner was rapidly increased from around 20 to 40 ºC at *Shangmei* phase, dropped to 30 ºC at *Liangmei* phase, and then increased gradually until reached to maximal 52 ºC at *Dahuo* phase, finally decreased to original temperature (25 ºC). The moisture was decreased from around 45 % to around 10 % during successive phases of incubation. Based on these results, a microbiological regulation for the production of *Fen-Hongxin Daqu* is proposed.

Keywords: Chinese liquor starter; Chinese liquor; traditional fermented; food microbial.

1. INTRODUCTION

Fen-Daqu is a natural fermentation starter, especially for distilled Chinese *Fen* liquor and traditional Chinese *Fen* vinegar production. *Fen-Daqu* is prepared from barley and peas by five

steps: (i) Ingredients formulation; (ii) Grinding and mixing; (iii) Shaping; (iv) Incubation (about 1 month) and (v) Maturation (about 6 months). The incubation step is divided into seven phases: *Woqu*, *Shangmei*, *Liangmei*, *Chaohuo*, *Dahuo*, *Houhuo* and *Yangqu*, as described previously [1].

The production of *Fen-Daqu* is still the constitution of the traditional fermentation technology without artificial added microorganisms. It has been reported all microorganisms related to saccharification and fermentation in the starter are derived from materials and from environment [2]. Other reported showed that the microbial distribution on the surface of *Fen-Daqu* were among of the bacteria, Lactobacillales, Actinomycetales, while among the fungi such as *Saccharomycopsis and Issatchenkia* were found in both the surface layer and the interior of *Daqu* [3]. *Fen-Daqu* also contain various enzymes, including amylase, protease, lipase, cellulose [4] and other metabolites, degradation products, and important flavor compounds [5].

Temperature plays an important role in the production of *Daqu*. The production of *Daqu* involves specific time-temperature control schemes resulting in a succession of microorganisms and natural result of metabolism. But until now, most of *Daqu* production still relies on workers' experience. During the production of *Fen-Daqu* almost physicochemical parameters such as temperature, relative humidity, and moisture are detected by workers' experience, such as "hand like a thermometer"[6].

We hypothesize that there was a converging relationship between the physicochemical change and microbial amount in *Fen-Daqu* during the incubation and they could be reflected the specific fermentation events and also relative to the quality of *Daqu*. But up to now, no microbial and physicochemical characteristics of *Fen-Daqu* during its phases of incubation have been reported.

The objective of this research was to determine microbiological and physicochemical changes during the incubation of *Fen-Daqu* and also to assess whether these parameters could be used to control the quality of *Fen-Daqu* intermediate products.

2. MATERIALS AND METHODS

2.1. Sampling

Fen-Daqu samples were obtained from Xinghuacun Fenjiu Group, Shanxi province, China. *Daqu* is fermented and matured in stacked layers. Samples were collected at the end of *Woqu*, *Shangmei*, *Liangmei*, *Chaohuo*, *Dahuo*, *Houhuo* and *Yangqu* phases. Each sample was obtained by randomly selecting from each upper, middle and lower stacked layer and mixed together as an experimental sample. Samples were stored at 4 °C until used.

2.2. Microbiological analysis

The samples were subjected to a microbiological analysis to monitor the change in the population during the incubation of *Fen-Daqu*. 10 grams of each sample was transferred into a sterile stomacher bag, 90 mL of saline-peptone water (8 g NaCl per liter, 1 g of neutral peptone per liter) was added, and the mixture was treated for 1.5 min in a stomacher machine. Subsequent decimal dilutions were prepared with the same diluents, and in all cases, duplicate counting plates were prepared of appropriate dilutions. After incubation, the colonies appearing on the selected plates were counted and calculated as colony-forming unit (cfu) per gram of *Fen-*

Daqu. All counts were repeated three times for each sample and results were reported as the means.

2.2.1. Total count of mesophilic aerobic bacteria (TMAB)

TMAB was enumerated in pour-plate of Plate count agar (PCA, Oxoid), incubation at 30 ºC for 48 to 72 h. Thermophiles were incubated for 24 ± 4 h at 55 °C.

2.2.2. Enterobacteriaceae

Selective enumeration was carried out in pour-plates of Violet Red Bile Glucose agar (VABG, Oxoid) with overlay with further VRBGA to cover the surface, after incubation at 37 °C for 24 ± 2 h.

2.2.3. Lactic acid bacteria (LAB)

LAB were enumerated in pour-plates of de Man, Rogosa and Sharpe medium (MRS, Oxiod) agar containing nystatin (1%), after incubation at 30 °C for 72 h.

2.2.4. Bacterial endospore

For the enumeration of bacterial endospore, 10% (w/v) sample suspension was heated at 80 °C for 5 min, suitably diluted, and spread on PCA plates, and then a top layer of 1.5 % agar was applied to restrict colony size and incubated at 30 °C for 48 to 72 h.

2.2.5. Fungi (Yeasts and molds)

Fungi were enumerated by pour-plates using Malt Extract Agar (MEA, Oxoid) and incubation at 37 ºC for 3 to 5 days.

2.3. Physiochemical analysis

2.3.1. pH measurements

Potentionmetric measurements of pH were carried out with a pin electrode of pH meter (PB-10, Sartorius, Germany) inserted directly into the sample. Three independent measurements were done on each sample. Means and standard deviations were calculated.

2.3.2. Determination of total acid

Acidity content of samples was determined in solution containing 25 g of *Fen-Daqu* in 150 mL of CO_2-free distilled water that was tritated with a standard NaOH solution approximately 0.1 N. Total titratable acidity was expressed as g lactic acid per kg dry matter [7]. Means and standard deviations were calculated on all data.

2.3.3. Determination of relative humidity and temperature in incubation room

The relative humidity and temperature in incubation room during the incubation of *Fen-Daqu* were simultaneously recorded via humidity/temperature logger (testo 175-H2). Means and

standard deviations were calculated.

2.3.4. Detection of temperature in incubation room

The temperature in incubation room was detected at the end of each phase by mini infrared thermometer gun (UNI-T UT301A). Means and standard deviations were calculated.

2.3.5. Detection of temperature in Daqu inner

The temperature in *Daqu* inner was recorded by ibutton (temperature sensor). In *Woqu* phase, randomly select 3 *Daqu* blocks for each incubation room, and mark them in different codes to avoid confusion. The ibutton was connected with computer by USB port, and then set 0 as starting point and one hour as duration for data recording. After that, it was sealed in a small plastic bag to protect it against corrosion. The ibutton was inserted in the inner of *Daqu* block, and then temperature was recorded until the end of *Yangqu* phase. Means and standard deviations were calculated.

2.3.6. Determination of moisture in Fen-Daqu

The moisture of the samples were determined by oven drying at 105 ºC until the weight remains constant [8]. The experiment was conducted in triplicate and the mean value determined.

3. RESULTS AND DISCUSSIONS

3.1. Study of microbial changes during the incubation of *Fen-Daqu*

Figure 1 showed that the level of total bacteria (incubated at 30 °C) rapidly increased during the early phases, after that reduced at *Chaohuo* phase and reach to the maximum level at *Dahuo* phase was around 11 log cfu/g and then gradually decreased over the later phases. This should be corresponding to the characteristic of temperature, relative humidity, and moisture profile (as showed in figure 4, 6 and 7) and the changes of mesophilic bacteria in these phases.

Figure 1 showed that the level of mesophilic bacteria (incubated at 55 °C) increased during the first and middle phases, attaining maximun level at *Dahuo* phase and relative decreased in the last phase. That could be explained by the increasing of temperature during the first and middle phases, which provided a proper condition for the growth of mesophilic bacteria (as showed in figure 6) and loss of moisture (as showed in see figure 7) within high temperature during the last phase could be attributed to the death of some microorganisms.

It was observed that the level of bacterial endospores (incubated at 30 and 55 °C) was relatively increased during the incubation of *Fen-Daqu* and attaining maximum level (around 9 log cfu/g) at *Yangqu* phase, as showed in figure 1. It could be explained that under unfavorable environmental conditions such as high temperature, low relative humidity and moisture, bacteria produced endospores.

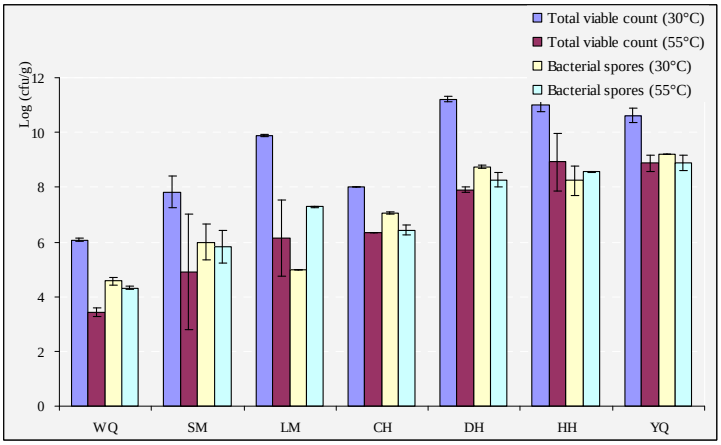

Figure 1. Change of vegetative cells and bacterial endospore during the incubation of *Fen-Daqu* (WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

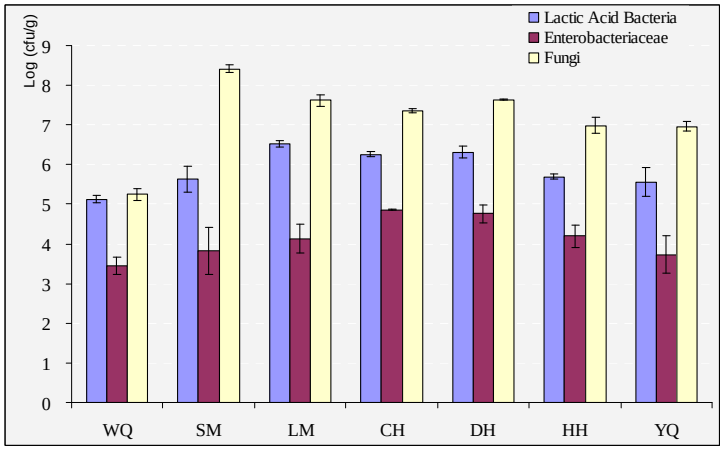

Figure 2. Change of LAB, enterobacteriaceae and fungi during the incubation of *Fen-Daqu* (WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

The total number of lactic acid bacteria, enterobacteriacea and fungi in *Fen-Daqu* during the incubation were showed in figure 2. It was observed that the level of LAB in the middle phases were relatively higher than in the first and later phases, and attaining maximum level at *Liangmei* phase. It should be related with the concentration of lactate, which is produced by LAB [9]. LAB plays an important role in food fermentation, cause the characteristic flavor changes associated with fermentation, it's also called as an efficient cell factory for food ingredient production [10]. Lactic acid bacteria were found in *Daqu*, such as *Weissella cibaria, Lactobacillus panis*, *L. helveticus*, *L. fermentum*, *L. pontis* [11].

The level of enterobacteriacea gradually increased during the first phases, and attaining maximum level (near 5 log cfu/g) at *Chaohuo* phase and then decreased during the later phases.

The level of fungi rapidly increased during the first phases, and attaining maximum level (> 8 log cfu/g) at *Shangmei* phase and then gradually decreased over the later phases. It could be explained that in the *Shangmei* phase reached to the optimum temperature, relative humidity and

moisture (as showed in figures 4 and 7) for fungal growth, therefore highest number were found. It was reported that the non-*Saccharomyces* yeasts represented most of the total yeasts population in *Fen-Daqu* [3], the non-*Saccharomyces* yeasts, such as *Trichosporon asahii*, *Debaryomyces hansenii*, *Hanseniaspora guilliermondii* were found in other *Daqu* [11]. They produce secondary metabolites, which can contribute to the final taste and flavor of wine [12]. Other reported showed that three types of mold (*Thermomyces*, *Penicillium* and *Aspergillus*) were found in *Fen-Daqu*. Among of them, *Thermomyces* were abundant in the interior *Daqu* [3]. That could be due to a higher temperature in the inner *Daqu* (as showed in figures 5 and 6).

Figures 1 and 2 showed that at *Woqu* phase, the total viable counts of bacteria, fungal and LBA counts were quite high (range of 5 - 9 Log cfu/g). It could be explained that most of them are derived from materials or environment [2]. In addition the incubation room often used for several batches of *Daqu* making without sterilization, therefore the spores accumulated in environment and bring to this phase.

It also observed that the level of fungi and LAB were lower than bacteria, which imply the dominant group of microorganism in *Daqu* is bacteria rather than fungi or LAB. That showed a positive correlation with the composition of microorganisms in *Daqu* [4, 13].

3.2. Physicochemical changes during the incubation

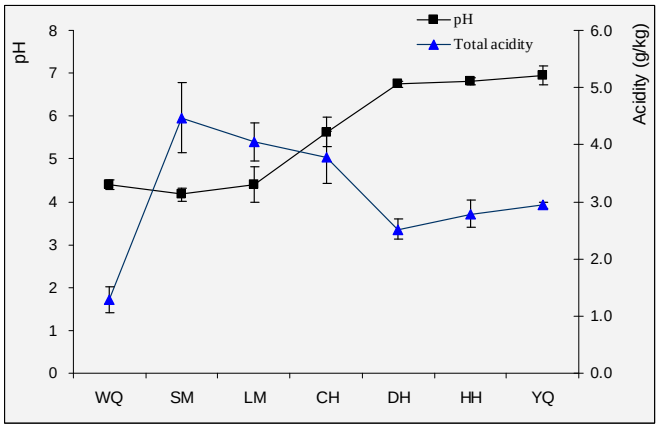

Figure 3. Change of pH and total acidity during the incubation of *Fen-Daqu*

(WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

Figure 3 showed that the pH increased over time during the incubation of *Daqu*. The rate of pH increase was slower in the first three phases (*Woqu*, *Shangmei* and *Liangmei*), and then become significant faster until *Dahuo* phase and finally keep in a steady level.

Figure 3 showed that the total acidity increased at first phases, attaining to the maximum level (near 5 g/kg) at *Shangmei* phase, after that decreased during the middle phases and then gradually increased again during the later phases. The total acidity in *Daqu* is derived from acid producing microbial species, which mainly produce acetic acid, lactic acid, or the degradation of lipid and protein, etc [14].

It was observed that the total acidity attain maximum level at *Shangmei* phase, while the maximal level of LAB occurred in *Liangmei* phase. It could be explained that other bacteria such as acetic acid bacteria also present with high number in *Daqu*, they produce acetic acid and will

lead increase of titratable acidity.

Figure 4 showed the changes of relative humidity and temperature in incubation room during the incubation of *Fen-Daqu*. The relative humidity was increased during the first phase and then decreased during later phases of the incubation. It attain at maximum level at *Shangmei* phase, this can be explained as that during this phase the temperature increased quickly as well as growth of microorganisms and the vapor released to the environment without artificial air ventilation. In other phases, since natural ventilation through turn of *Daqu* and open air windows and doors the relative humidity was reduced from 100 % to 20 %. It was also observed that the temperature increased rapidly in *Shangmei* phase from 15 ºC up to 40 ºC and *Chaohuo*, *Dahuo* phases attain maximum 45 ºC. That showed a positive correlation with the change of microbial count during these phases (see figures 1 and 2). In *Liangmei* phase, due to the good ventilation the heat was released to the surroundings at a lower temperature about 20 ºC, that in order to prevent damage overheating.

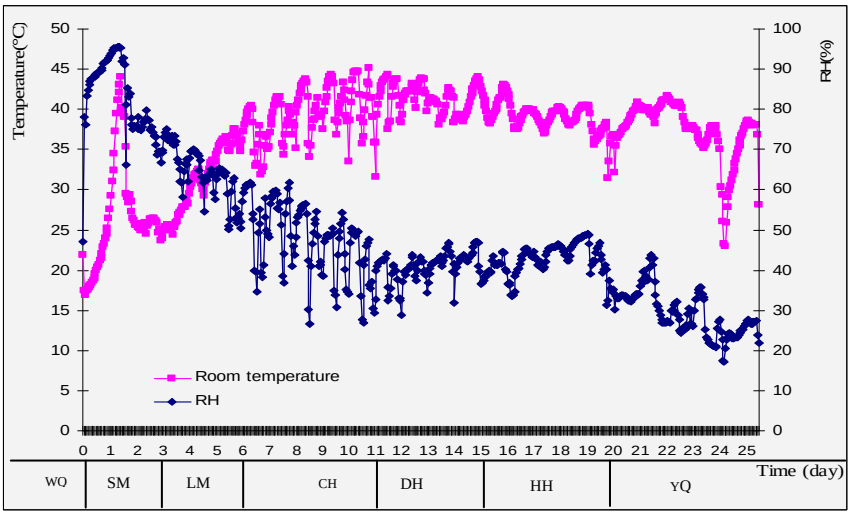

Figure 4. Change of temperature and RH in incubation room during the incubation of *Fen-Daqu* (WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

Figure 5 showed that the temperature in surface of *Daqu* was increased from the end of *Woqu* phase to the end of *Dahuo* phase, and then reduced during the later phase. It could be related to the growth of microbial during these phases.

Figure 6 showed that the room temperature increased over time during production of *Daqu*. Exception of *Yangqu* phase, the inner temperature of *Daqu* was higher than room temperature, that's mainly because of microbial growth in *Daqu*.

During the incubation of *Daqu*, at the beginning of *Shangmei* phase, the inner temperature decreased from 25 ºC to 18 ºC, and then rapidly increased to a higher level of above 40 ºC. After that, it dropped to 33 ºC at the end of *Shangmei* phase. During *Liangmei* phase, the temperature decreased again to 30 ºC and increased gradually until reached to maximal 52 ºC at the beginning of *Dahuo* phase. From that level, the temperature started to decrease slowly and finally back to original temperature (25 ºC). The aim of *Shangmei* to *Liangmei* phase is to activate initial microbial growth and to allow the temperature to increase gradually, attaining 30-40 ºC in 3-5 d. The initial 24-48 h is considered as a crucial time for establishing the structure of *Daqu*'s microbial community, and hence the pioneer microorganisms such as fungi start to

colonize and mycelium will spread over the surface of *Daqu* [1].

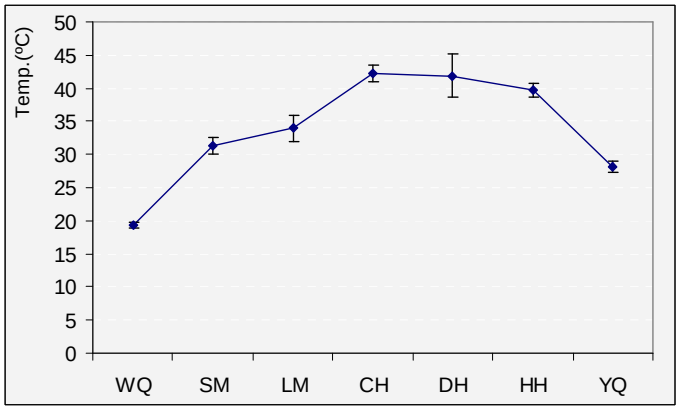

Figure 5. Change of temperature in incubation room

(WQ: *Woqu*, SM: *Shangmei*, LM: *Liangmei*, CH: *Chaohuo*, DH: *Dahuo*, HH: *Houhuo*, YQ: *Yangqu*).

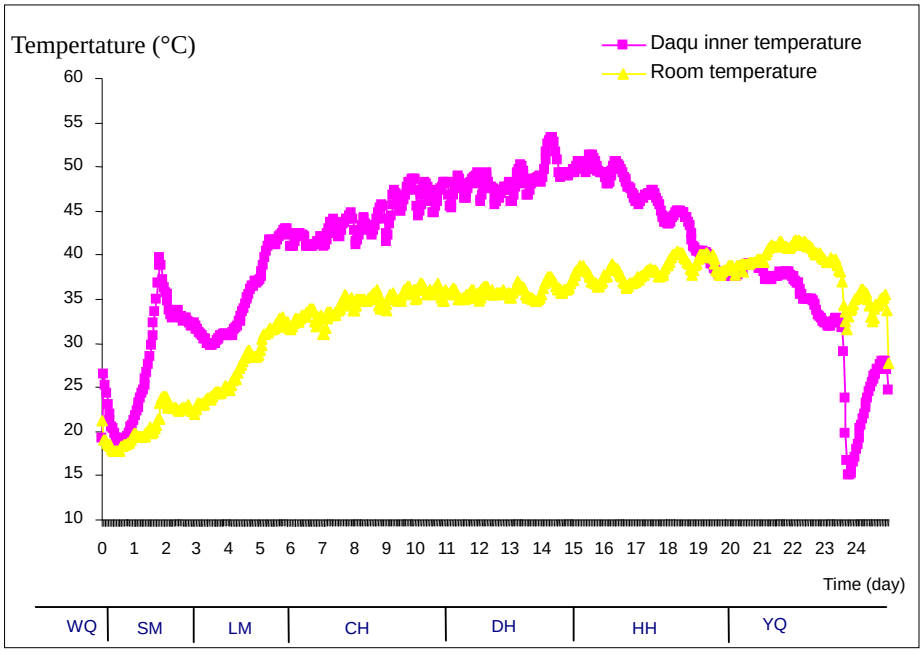

Figure 6. Change of temperature in *Daqu* inner and incubation room during the incubation of *Fen-Daqu*.

(WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

High temperature during the incubation phase (*Chaohuo*, *Dahuo* and *Houhuo*) could enhanced proteolysis and accumulation of amino acids [15, 16], and help to produce more volatile compounds such as pyrazines that could be formed through the Maillard reaction between saccharides and amino residues [17, 18].

The room temperature measured by IR thermometer (figure 5) was significant different with the data obtained with ibutton (figure 6), however the general trend is quite similar. Since

IR thermometer was placed in the space between *Daqu* blocks, and the distance to *Daqu* is quite short, therefore the measurements easily can be influenced by the activity inside of *Daqu*, especially the growth of different microorganisms. The ibutton was placed on the wall, which gives more accurate results and reflect the changes of room temperature.

Figure 7 showed that the moisture in *Fen-Daqu* samples was decreased from around 45 % to around 10 % during successive phases of incubation. During these phases the moisture rather rapid decreased from *Shangmei* to *Houhuo* phase, due to the increased temperature and decreased of relative humidity in incubation room (as showed in figure 4), and good ventilated.

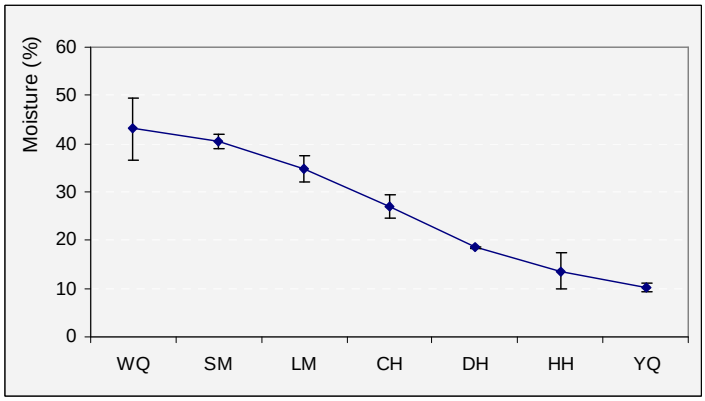

Figure 7. Change of moisture in *Fen-Daqu* during the incubation.

(WQ: *Woqu*; SM: *Shangmei*; LM: *Liangmei*; CH: *Chaohuo*; DH: *Dahuo*; HH: *Houhuo*; YQ: *Yangqu*).

The moisture in *Woqu* phase showed a positive correlation with the percentage of water was added to the grinding and mixing stage about 40 %. There was a gradual decrease in the moisture content of the samples from *Houho* to *Yangqu* phase. That showed a positive correlation with the aim of this phase is to allow the equilibration of moisture, acidy and enzyme activity [16].

4. CONCLUSION

The microbial and physiochemical changes in *Fen-Daqu* during the incubation were determined in this study. It also revealed a strong correlation between microbial and physiochemical measurements. This could help *Daqu* producers to monitor the progress of the *Daqu* manufacturing process by measuring the physiochemical parameters, in order to regulate the functional strains.

Acknowledgment. We thank Shanxi Xinghuacun Fenjiu Group Company for *Daqu* sampling assistance and advice. This study is funded by National Natural Science Foundation of China (No. 31071592) and KNAW-China Joint Research Project (No. 07CDP015) from the Royal Netherlands Academy of Arts and Sciences.

REFERENCES

1. Zheng X. W., Tabrizi M. R., Nout M. J. R. and Han, B.-Z. - *Daqu* – A traditional Chinese liquor fermentation starter. Journal of the Institute of Brewing **117** (2011) 82-90.

2. Xiong Z. S. - Research on three flavor type liquors in china (III) Fen-flavor Liquor-Xinghuacun, Liquor-Making Science & Technology **133** (2005) 17–21.
3. Shi J. H., Xiao Y. P., Li X. R., Ma E. B., Du X. W., Quan, Z. X. - Analyses of microbial consortia in the starter of *Fen* Liquor, Letters in Applied Microbiology **48** (2009) 478-485.
4. Li C., Mu L., Wang J. Y., Lei Z. H., Chen J. Y., Han, B. Z. - Physiochemical and microbiological analysis of *Fen*-type *Daqu*, China Brewing **1** (2009) 140-142 (in Chinese).
5. Wu X. H., Zheng X. W., Han B. Z., Vervoort J., Nout M. J. R. - Characterization of Chinese liquor starter, "*Daqu*", by flavor type with ^{1}H NMR-based nontargeted analysis,. Journal of Agricultural and Food Chemistry **57** (2009) 11354-11359.
6. Di H., Zhang R. P., Yang Z. J. - Discussion on the improvement of the production of *Fen-Daqu*. Shanxi Food Industry **01** (1995)(in Chinese).
7. Hou M. L. - Food analysis. Chemical Industry Press Beijing, 2004.
8. AOAC, Official methods of analysis (15th ed.), In Association of Official Analytical Chemists: Washington, DC, 1990.
9. Liu S. Q. - Practical implications of lactate and pyruvate metabolism by lactic acid bacteria in food and beverage fermentations, International Journal of Food Microbiology **83** (2003) 115-131.
10. Hugenholtz J. - The lactic acid bacterium as a cell factory for food ingredient production, International Dairy Journal **18** (2008) 466-475.
11. Gao Y. B. - Investigation of microbial community of Chinese liquor *Daqu* by polymerase chain reaction-denaturing gradient gel electrophoresis. Jiangnan University, Master thesis 2010 (in Chinese).
12. Braulio E. Z., Manzanares P., Ramön D., Quero A. - The role of non-Saccharomyces yeasts in industrial winemaking, International Microbiology **1** (1998) 143-148.
13. Wang C. L., Shi D. J., Gong G. L. - Microorganisms in *Daqu*: a starter culture of Chinese Maotai-flavor liquor, World Journal of Microbiology and Biotechnology **24** (2008) 2183-2190.
14. Shen C. H., Hong Y., Xu D. F. - Study on *Daqu* Quality Standards (IV) —Investigation on Physiochemical Property Indexes of *Daqu*. Liquor-making Science & Technology **9** (2005) (in Chinese).
15. El-Ella W. M. A., Baky A. A. A., Aly M. E., Fox P. F. - Effect of ripening temperatures on proteolysis and lipolysis in the outer and inner regions of Ras-type cheese made by various salting methods, Food chemistry **28** (1988) 1-16.
16. Zheng X. W., Tabrizi M. R., Nout M. J. R., Han B. Z. - *Daqu* – A traditional Chinese liquor fermentation starter, Journal of the Institute of Brewing **117**(2011) 82-90.
17. Owens J. D., Allagheny N., Kipping G., Ames J. M. - Formation of volatile compounds during Bacillus subtilis fermentation of soya beans, Journal of the Science of Food and Agriculture **74** (1997) 132-140.
18. López-Galilea I., Fournier N., Cid C., Guichard E. - Changes in headspace volatile concentrations of coffee brews caused by the roasting process and the brewing procedure, Journal of Agricultural and Food Chemistry **54** (2006) 8560-8566.

TÓM TẮT

NGHIÊN CỨU SỰ THAY ĐỔI CỦA VI SINH VẬT VÀ YẾU TỐ LÍ HÓA TRONG QUÁ TRÌNH SẢN XUẤT BÁNH MEN RƯỢU PHẦN

Lê Văn Điệp[1,*], Han Bei-Zhong[2]

[1]*Khoa Hóa học, Trường Đại học Vinh, 182 Lê Duẫn, TP. Vinh, Nghệ An.*

[2] *Khoa Công nghệ Thực phẩm, Trường Đại học Nông Nghiệp Trung Quốc.*

[*]Email: *levandiep@vinhuni.edu.vn*

Bánh men rượu Phần (*Fen-Daqu*) vừa là tác nhân đường hóa, vừa là tác nhân lên men trong sản xuất rượu Phần, một loại rượu trắng có hương thanh nhẹ nổi tiếng ở tỉnh Sơn Tây, Trung Quốc. Nó được sản xuất bởi quá trình lên men tự nhiên từ nguồn nguyên liệu đại mạch và đậu Hà lan. Phương pháp nuôi cấy vi sinh vật truyền thống và các kĩ thuật phân tích trực tuyến được sử dụng trong phân tích sự thay đổi của các nhóm vi sinh vật và các yếu tố lí hóa.

Sự thay đổi của vi sinh vật phân biệt theo từng nhóm hiếu khí (30 °C), vi sinh vật ưa nhiệt (55 °C), bào tử vi khuẩn (30 °C và 55 °C), vi khuẩn lactic, enterobacteriaceae và nấm từ mức thấp nhất khoảng 10^6, 10^4, 10^5, 10^5, 10^5, 10^4 and 10^5 cfu/g và đạt cực đại ở khoảng 10^{11}, 10^9, 10^9, 10^9, 10^7, 10^5 and 10^8 cfu/g. Trong quá trình ủ vi sinh vật thay đổi theo xu thế tăng dần từ *Woqu* đến *Liangmei* và sau đó giảm dần theo thời gian. pH tăng dần, còn độ ẩm phòng giảm dần từ khoảng 100 % xuống còn 20 % và thủy phần bánh men giảm dần từ khoảng 45 % xuống còn khoảng 10 % theo thời gian quá trình ủ. Nhiệt độ phòng ủ tăng dần từ pha đầu cho đến pha giữa và giảm nhẹ ở pha sau. Nhiệt độ bên trong bánh men tăng nhanh ở giai đoạn *Shangmei* đạt 40 ºC sau đó giảm xuống còn 30 ºC ở giai đoạn *Liangmei*, tiếp đó tăng và đạt đến cực đại 52 ºC tại *Dahuo*, cuối cùng giảm dần đến nhiệt độ phòng (25 ºC) ở pha sau.

Từ các kết quả trên và qua phân tích sự tác động qua lại giữa chúng có thể ứng dụng vào quá trình điều chỉnh sự phát triển của vi sinh vật trong quá trình sản xuất bánh men.

Từ khóa: bánh men rượu, rượu trắng, lên men truyền thống, vi sinh vật.