

The natural microflora of Xuanwei ham and the no-mouldy ham production

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Abstract

The natural microflora of Xuanwei ham in China was investigated and the results showed that there was a close relationship between yeast and ham quality. As the dominant microorganism, ham yeast played an important role in the quality of ham. Other mycelium fungi such as *Penicillium* and *Aspergillus*, which looked flourishing on the surface of ham, may not avail ham quality because they produce some mycotoxins. The quality of ham inoculated with yeast naturally isolated from ham was superior to that of traditional ones according to indices of color–fragrance–flavor and the contents of amino acids. By inoculating yeasts on ham and controlling the air humidity of ham room, a better quality ham called no-mouldy ham was developed successfully.

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1. Introduction

Xuanwei ham, one of the three famous hams in China, is produced in Xuanwei district of Yunnan province with a history of more than a 1000 years. The yield of Xuanwei ham is more than 10,000 tonnes per year. Many microorganisms grow in or on traditional Xuanwei ham (Fig. 7). However, few literatures could be found about studies on the microorganisms on Xuanwei ham.

Moulds are usually thought as dominant microorganisms on the surface of ham that avail to ham quality. Many microbiological investigations on western ham have been done. Moulds are considered helpful to the ripening of dry-cured meat products due to their positive effects on flavor and appearance (Lücke, 1986). *Staphylococcus* and *Micrococcus* may not be recovered at the end of maturation of Iberian dry-cured ham, whereas yeasts are the predominant microorganism

(Rodríguez et al., 1994). The superficial yeast population of Iberian ham is shown to be useful for estimating the progress of maturation. *Candida zeylanoides* is the dominating yeast in the early stages, and *Debaryomyces hansenii* is the dominating one in the matured hams (Núñez, Rodríguez, Córdoba, Bermúdez, & Asensio, 1996). Yeasts are suggested to be helpful to curing process and may make desirable effects through their proteolytic or lipolytic activity (Saldanha-da-Gama, Malfeito-Ferreira, & Loureiro, 1997).

Mycotoxins, which include aflatoxin, ochratoxin, and zearalenone, etc., may develop on various foods and feeds from the secondary metabolism of some filamentous fungi or moulds such as *Aspergillus*, *Penicillium*, *Trichoderma*, *Alternaria*, and *Fusarium*. Mycotoxins cannot be destroyed by normal industrial processing or cooking because they are heat-stable. They could cause serious risks for human and animal health (Brera, Miraglia, & Colatosti, 1998). Most moulds are toxigenic when tested with brine shrimp larvae and VERO cells for mutagenicity in the Ames test and for antimicrobial activity against *Staphylococcus aureus*. The toxigenic

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potential population increases as the processing progress (Cvetnić & Pepeljnjak, 1997; Escher, Koehler, & Ayres, 1973; Núñez, Rodríguez, Bermúdez, Córdoba, & Asensio, 1996; Wu, Ayres, & Koehler, 1974). Treatments to prevent the growth of undesirable moulds would also prevent the growth of beneficial ones. The use of non-toxicogenic strains as starters could help to minimize health hazards associated with Iberian ham, without affecting the quality and character of the product (Escher et al., 1973; Wu et al., 1974).

To determine the role of microorganisms that grow on or in ham, we surveyed the microflora on Xuanwei ham and studied the roles of each microorganism to ham quality. Through our studies on Xuanwei ham for more than 5 years, we discovered that ham yeasts are the dominate microorganism in ham and play an important role during whole fermentation period. The amount of ham yeast was about $10\text{--}10^3$ times more than that of other microorganisms. The quality of ham could be improved by inoculating yeast on ham.

2. Materials and methods

2.1. Procedure of ham processing

Usually, the pigs called Wujin breed, which was bred in Xuanwei district, were slaughtered annually on Spring Festival corresponding to February. Then the legs (8–12 kg for each) were cut off from the body and cooled for about 10 h in room temperature until the meat temperature dropped to 5–6 °C. The legs were then trimmed neatly. Salt (2 kg/100 kg ham) was brushed on hams for first time, and the blood liquid was extruded from vessel thoroughly by hand. After 2 days, salt (4–5 kg/100 kg ham) was put on hams for the second time and the liquid was extruded further. Salt (1–2 kg/100 kg ham) was put on hams for the third time after 2 more days, and this step was supplementary to some hams if they did not have enough salt on them, which was judged objectively. Then the hams were piled up and the liquid was removed naturally by gravity. After half a month, the hams were hanged in rows with ventilation on sunny day but not on rainy day. The hams will mature primarily at Duanwu festival corresponding to June and mature thoroughly at Medium Autumn festival corresponding to September. The no-mouldy ham procedure was inoculating yeast mixture at salting time by mixing the yeast culture in salt. The humidity of ham house was kept lower for this procedure.

2.2. Isolation of microorganism

Five hundred Xuanwei hams were employed in the procedure of isolating fungi, bacteria, and actinomycetes. The media used in the experiments were potato-

dextrose agar, Czapek agar, Martin Bennett's agar, maltose extract agar, and nutrient agar (Fan, Li, & Shen, 1989). Ten percent of slat was added in half of the media. Half of the agar plates were incubated at 25 °C and the other half incubated at 5 °C.

For isolating microorganisms, samples were fetched from the surface of hams and underneath 2 cm in hams with an aseptic sample-fetching appliance (1 cm diameter). Samples were taken at three different positions of each ham. The samples were immediately homogenized in sterile PBS solution in a sterile blender cup under aseptic condition. Various microorganisms from ham were separated and counted by diluted plate (Kondo & Kato, 1983). Totally 3400 microorganism strains were isolated and the identification was done on the basis of macro and microscopical characteristics, sporulation mode, and some biochemical tests. The cultures were maintained at 4 °C on slants of the same media. Microorganism strains with distinct characters were screened out for further experiments.

2.3. The determination of microorganism population in the air

Aerobiological investigation of microorganism was done according to Su et al. (2002). A total of 200 plates were examined, and the identification and counts of air microorganisms was done as described in Section 2.2.

2.4. Reversed inoculation test

Every strain isolated from the homogenate was cultured in potato-dextrose liquid culture containing 10% NaCl at 25 °C for 7 days with shaking (700 rpm). The culture was then mixed with salt for inoculating on ham at salting time. The inoculum was 0.1% of ham weight. Each group consisted of 10 legs, which were selected randomly. The legs were then processed according to traditional procedure.

2.5. Determination of sensory quality of ham

The quality of ham was identified by 15 specialists. After discussing by specialists panel, the score was distributed as: odor 40, tastes 40, and color 20 (GB18357-2001).

2.6. Toxic substance analysis

Samples were took from the surface of matured hams, which were inoculated reversely with strains of *A. flavus*, *A. fumigatus*, *A. versicolor*, and some typical and high frequency appearance strains in the genera of *Penicillium* and *Fusarium*, which were isolated from ham previously. Qualitative determination of aflatoxins, ochratoxin, T-2, and deoxinivalenol (DON) was carried

out by using Lin's method (1998). Identification of sterigmatocystin was done by following the method of Stack and Rodricks (1971). Identification of patulin and penicillic acid was done according to the method of Erdogan, Gurses, and Sert (2003).

2.7. Analysis of amino acid

Three mg of dry ham meat was placed in an ampoule containing 1 ml of 1 N HCl. The sealed ampoule was kept at 100 °C in an oven for 18 h. After cooled, the hydrolysate was filtered through Whatman No. 1 filter paper according to the method in Food Chemical Analysis (Huang, Zhao, Lai, Mao, & Wei, 1978). Samples of amino acid were analyzed by the HITACHI 835–50 type amino acid automatic analysis apparatus.

3. Results and discussion

3.1. Natural microflora of Xuanwei ham

Actinomycetes strains in 7 groups were acquired. The dominant strains were in *Streptomyces* that accounted for almost half of the isolated actinomycetes strains. Actinomycetes and bacteria were not the dominant microorganism on ham (the detailed results shown in Fig. 1). While few literatures could be found about the effect of actinomycetes on ham quality, bacteria, except for some micrococci and lactobacteria, were usually thought to be harmful to human health and cause hams spoiled (De Martinis & Freitas, 2003; Losantos, Sanabria, Cornejo, & Carrascosa, 2000; Portocarrero, Newman, & Mikel, 2002; Rodríguez et al., 1994).

Aspergillus and *Penicillium* looked flourishing on the surface of ham in July and August. Eight species in *Aspergillus* was found and the dominant species was *A. fumigatus*, which account for one third of all *Aspergillus*. The less dominant species were *A. flavus* and *A. versicolor*. Other species occurred occasionally. Four species in *Penicillium* were found. *P. lanate*, *P.*

divaricate, and *P. velutin* were almost equally isolated, while *P. monovorticillate* appeared in a smaller proportion. Generally, high RH values are beneficial to the growth of moulds on the surface of ham (Arnau, Gou, & Comaposada, 2003). Some Moulds could improve the quality of ham, but many moulds on ham may produce mycotoxic substance (Creppy, 2002; Escher et al., 1973; Martín, Córdoba, Benito, Aranda, & Asensio, 2003).

Yeasts were the dominant fungi, including *Saccharomyces*, *Schizosaccharomyces*, *Hansenula*, and a special black yeast. *Saccharomyces*, *Schizosaccharomyces*, and *Hansenula* were recovered at the same frequency, and each accounted for one third of the microorganisms isolated. Black yeast appeared occasionally. We found that yeasts accounted for more than 50% of the total microorganisms on mature ham at any given time. The result was similar to that of other studies on different type of ham. The amount of yeasts from pork-based products, such as country-cured hams and several types of bacon, ranges from 10^3 – 10^9 cfu/g of fat. Some desirable effects appear to be related to their proteolytic or lipolytic activity (Deak, 1991; Saldanha-da-Gama et al., 1997). While *Staphylococcus* and *Micrococcus* could not be recovered at the end of maturation of Iberian dry-cured ham, yeasts are the predominant microorganism (Núñez et al., 1996; Rodríguez et al., 1994).

3.2. The annual variation of microflora

Usually, on a given medium, the growth mode of microorganism is decided by humidity and temperature to a great extent (Arnau et al., 2003). The rainy season in Xuanwei district is from April to September, during which the temperature and humidity were higher than those of the other season, called dry season (Figs. 2 and 3).

The annual variation of microorganisms on ham was shown in Fig. 4. *Penicillium* and *Aspergillus* varied with the humidity. They were rarely found in the inner part of ham. Their quantity increased in April when the temperature and humidity were higher and reached their peak

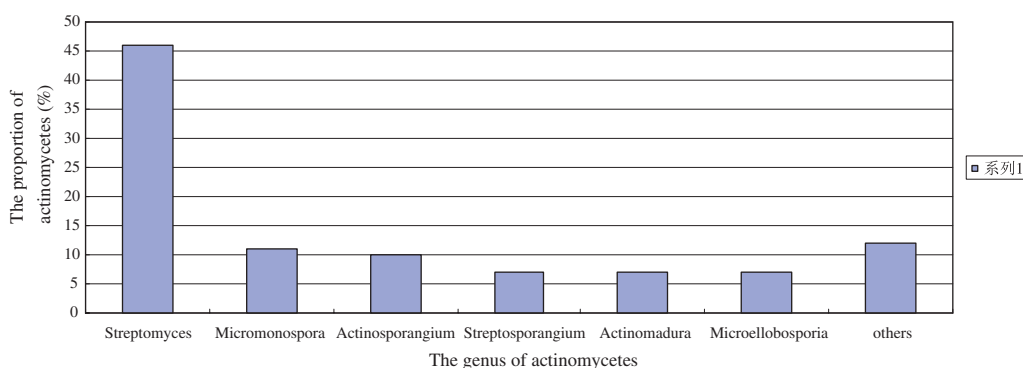


Fig. 1. The proportion of different isolated strains of actinomycetes.

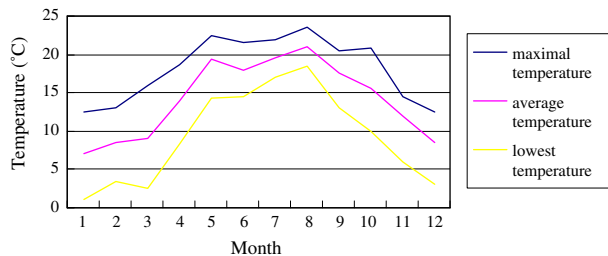


Fig. 2. Annual variation of temperature in ham storehouse.

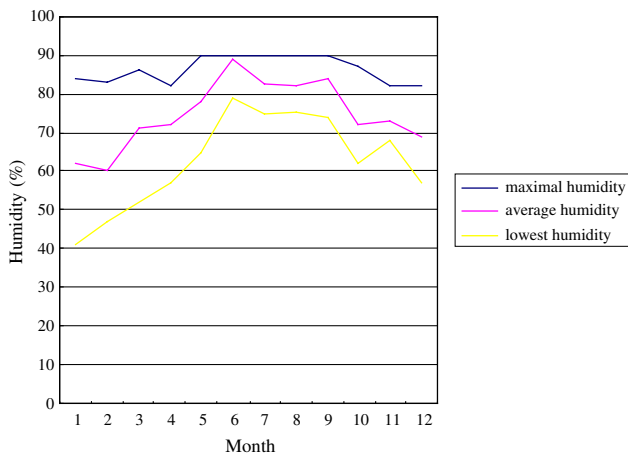
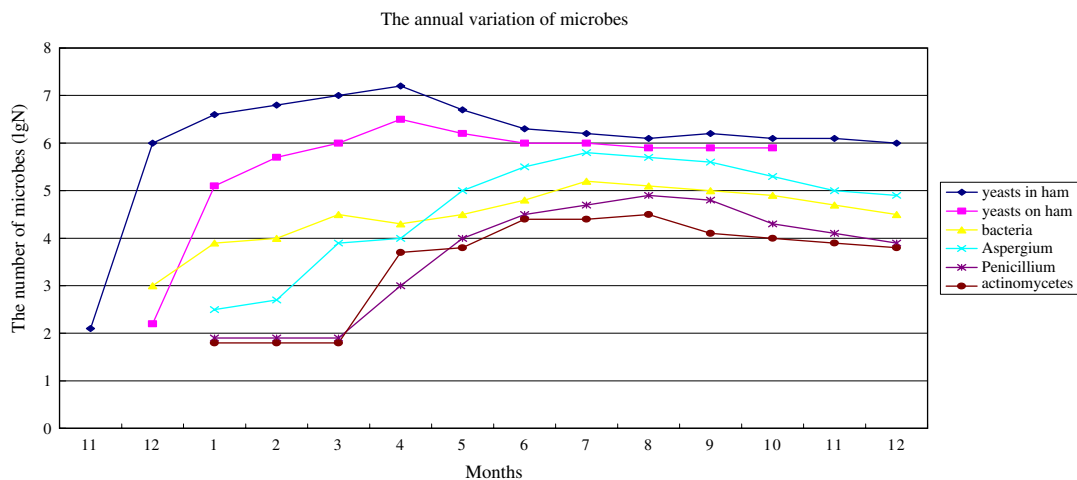


Fig. 3. Annual humidity variation in ham storehouse.

in July or August. The spores of *Aspergillus* reached 7×10^5 cfu/g in August, and that of *Penicillium* were 9×10^4 cfu/g at its peak. Generally, the growth of moulds such as *Penicillium* and *Aspergillus* were related to the temperature and humidity in ham room. Their growth conditions were: lowest temperature >15 °C, average temperature >20 °C, and average humidity

$>80\%$. *Mucor*, *Trichoderma*, and *Botrytis* were similar to *Aspergillus*, but they occurred in a smaller proportion. According to our observation, moulds started to grow in May and became flourishing in June. Their mycelium would lyse and the spores formed in August and September. The quantities of their spores fall off gradually after September. This suggested that mould may not play a role in some parts of the ham sometimes. The growth behavior of bacteria and actinomycetes did not depend on the humidity. The quantities of bacteria and actinomycetes varied from that of mould, and they were smaller than that of yeast. We thought that they may not be the functional microorganism. From the beginning of salting time to a month, yeasts on ham multiplied exponentially and reached their peak in April. Then the number dropped and became stable (2×10^7 cfu/g). Yeasts in ham varied accordingly. This showed that yeast can grow in all the part in the ham, which suggested that yeast may play an important role in ham fermentation.

According to the above data, yeasts were the natural dominant microflora in Xuanwei ham. They were the largest population and accounted for 60–70% of the total microorganism population on the surface of ham. From the beginning of salting time to April, the yeasts were absolutely predominant. The amount of yeasts could reach 1×10^6 cfu/g to 3×10^6 cfu/g in mature ham muscle. Sometimes the number of yeast was nearly 100 times more than that of *Aspergillus* and *Penicillium*. The second largest population was *Aspergillus* and *Penicillium*, and they account for 5–6%, respectively. Mould did not multiply until April, and they flourish from May to July when the temperature and humidity were higher. Even at that time, the amount of yeast were 3–30 times as much as that of *Aspergillus* and *Penicillium*. A condition of RH $<60\%$ might inhibit mould growth. So, it is recommended that high RH values

Fig. 4. Annual variation of microorganism community of Xuanwei ham: (a) yeasts on ham; (b) yeasts in ham; (c) *Aspergillus*; (d) *Penicillium*; (e) bacteria; (f) actinomycetes.

are avoided during resting in order to prevent undesirable moulds (Arnaud et al., 2003). Sometimes, a few hams with good quality did not have mould growing on their surface at all, but the amount of yeast was equal to that of mouldy ham. Thus, we speculate that yeast may be the major functional microorganism to ham fermentation.

3.3. The annual variation of microorganisms in the air of ham room

The variation of microorganism population in the air of ham house differs from that on the ham (Fig. 5). Yeasts in the air appeared 2 months after salting (500 M^{-3}), and the number was steady until June. In July, the amount of yeast increased dramatically and it continued increasing until September ($4.8 \times 10^4 \text{ M}^{-3}$). After October, the

amount dropped rapidly. The spores of *Aspergillus* in ham house increased in April until reaching its peak in August. *Penicillium* was found in March and reached its peak in October ($2 \times 10^4 \text{ M}^{-3}$). The appearance of the peak number of all kinds of microorganisms was 2 months later than that of the corresponding microorganisms on ham. Thus, the microorganism in the air might originate from hams, and obviously varied with the temperature and humidity of ham house.

3.4. Reverse inoculation on ham by microorganism

Strains of fungi and yeast with distinct characters were selected from all of the isolates for the test to validate the role of different microorganisms. The detailed results were shown in Table 1. Seventy-two strains

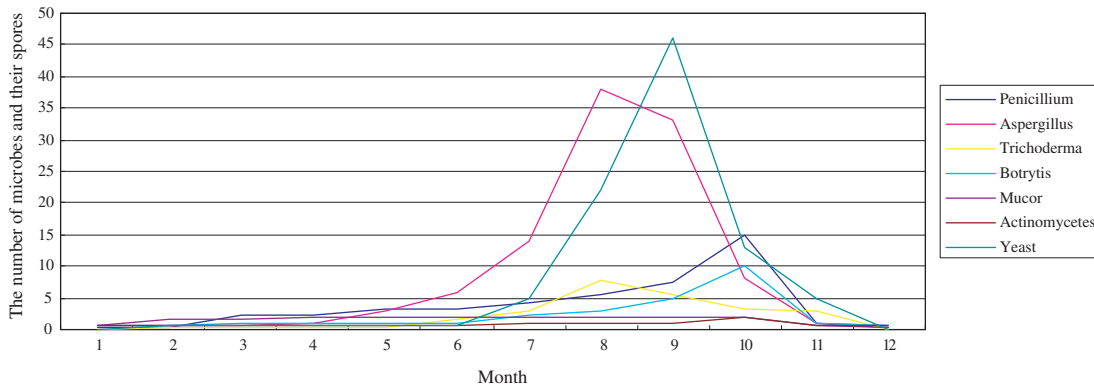


Fig. 5. Annual microbe variation in ham storehouse space.

Table 1
The selected fungi strains on Xuanwei ham for reverse inoculation experiments

Genus or group of isolated microorganisms	Strain number	Genus or group of isolated microorganisms	Strain number
<i>Aspergillus</i>			
<i>A. fumigatus</i>	8	<i>Trichoderma</i>	4
<i>A. flavus</i>	5	<i>Rhizopus</i>	3
<i>A. versicolor</i>	5	<i>Mucor</i>	2
<i>A. niger</i>	2	<i>Paecilomyces</i>	2
<i>A. ochraceus</i>	2	<i>Helminthosporium</i>	2
<i>A. wentii</i>	1	<i>Alternaria</i>	2
<i>A. candidus</i>	1	<i>Fusarium</i>	2
<i>A. restrictus</i>	1	<i>Botrytis</i>	2
<i>Penicillium</i>			
<i>Monoverticillate</i>	1	<i>Clasterosporium</i>	1
<i>Ianate</i>	3	<i>Stremphylium</i>	1
<i>Divaricate</i>	4	<i>Ozonium</i>	1
<i>Velutina</i>	4	<i>Gliocladium</i>	1
<i>Yeast or yeast-like fungi</i>			
<i>Saccharomyces</i>	4		
<i>Schizosaccharomyces</i>	3		
<i>Hansenula</i>	4		
Black yeast	1		

Table 2
Toxic strains of *Aspergillus* and *Penicillium*

	<i>A. fumigatus</i>	<i>A. flavus</i>	<i>A. versicolor</i>	<i>Penicillium</i>	<i>Fusarium</i>
Total strains	8	5	5	12	2
Toxic strains	2	1	1	1	0

distributing in different groups or genera of fungi, such as *Aspergillus*, *Penicillium*, and *Trichoderma*, and 2 lactobacillus isolated from Xuanwei ham, were each inoculated reversely to 10 hams. When ham matured, the score of inoculated ham were lower than that of traditional ham except for hams inoculated with yeasts (data not shown). Thus, we thought that the fungi and lactobacillus were not the microorganisms that contribute to ham quality.

Reverse inoculation by strains isolated from ham showed that the yeast was the contributing microorganism to ham quality. Among 12 yeast strains inoculated, 2 strains, XY8621 and XY8662, were best to ham fermentation. These strains of ham yeasts were identified as *Hansenula xuanweiensis* Jiang sp. Nov. and *Hansenula anomala* var. *xuanweiensis* (Jiang, Ma, Wang, Zhang, & Jiang, 1994). The strain XY8621 possessed a certain activity of proteinase and strain XY8662 produced fragrant ester. This result was different from that of Núñez et al. (1996) and Saldanha-da-Gama et al. (1997), which show that the predominant yeast species on ham belong to *Debaryomyces*, *Cryptococcus*, *Pichia* genera. Thus, different type of hams may have their own fermentation yeast.



Fig. 6. No-mould ham.

3.5. Toxicity analysis of moulds

According to the traditional view, a good Xuanwei ham must have “green gown” on it, which means on the surface of ham there are flourishing green moulds. Some literatures implies that moulds may play good role in the fermentation of ham (Martín et al., 2003), while others showed that *Penicillium*, *Fusarium*, and *Aspergillus* are the major toxic fungi on food, such as dry-cured Iberian ham (Brera et al., 1998; Cvetnić & Pepeljnjak, 1997; Erdogan et al., 2003; Escher et al., 1973; Fields, 1979; Núñez et al., 1996). We found 15.6% of mould strains examined produced toxic substance. The toxic substance could penetrate underneath 0.6 cm of ham muscle (Table 2). Since many other strains and mycotoxins were not examined, more mycotoxin might be present in the ham. The result showed that moulds might not be beneficial to ham quality. Thus, we should avoid moulds growing on ham.

3.6. The production of no-mould ham

Since yeasts played an important role in ham quality while mould was not beneficial, we decided to develop a no-mould Xuanwei ham procedure. We inoculated the mixture of yeast strains XY8662 and XY8621 at salting on selected 600 legs (10.5 tonnes totally). The legs were then processed according to the traditional method except for keeping the humidity in ham house lower when ham ferment. The amount of yeasts in the inoculated



Fig. 7. Traditional ham.

hams were 3–5 times as that of the traditional ones within 1 month, and the time of reaching their peak number appeared 10 days ahead of traditional ones. The mature time of hams was shortened by 1–1.5 months (Fig. 6).

In addition to inoculating yeast on ham, keeping humidity less than 80% was another key point should be controlled in the development of no-mould ham. The major method was to keep window open and ventilate ham room on sunny days and to shut down window on rainy days.

The comparison of the quality of matured yeast-inoculating hams and traditional ones was done by 15 specialists according to GB18357-2001, *Xuanwei ham*. The total score was 100. Scores 20, 40, and 40 were given to indices of color, odor, and taste, respectively, which was decided by specialist panel (Table 3). The odor score of raw inoculation ham were 24.2% higher than that of traditional ham, and the odor score of cooked inoculation ham were 15.6% higher than that of traditional ham. The taste score of raw inoculation ham were 16.5% higher than that of traditional ham, and the taste score of cooked inoculation ham were 18.6% higher

than that of traditional ones. The color score of raw inoculation ham was 11.9% higher than that of traditional ham, but the color score of cooked inoculated ham dropped 13.1% lower than that of traditional ones, which was the only index in inoculation ham lower than that of the traditional ones. The reason for this was currently unknown. The total marks of inoculated ham were 14.4% higher than that of traditional ones. This showed that yeasts play an important role in ham quality (Fig. 7).

In addition to good quality, no-mould ham looked hygiene and had a high utility of muscle when processed further because less dirty surface of ham meat should be wiped off.

3.7. The effects inoculated yeast on amino acid of ham

Amino acids, peptides, inorganic salts, and nucleotides are the main taste compounds in meat products (MacLeod, 1996). Nucleotides decreased to undetectable levels during the curing process in sausages and Parma hams. Therefore, in dry-cured products that

Table 3
Comparison of scores for color, odor, and taste between inoculated and traditional ham

Treatment	Marks						Total
	Odor		Taste		Color		
	Raw	Cooked	Raw	Cooked	Raw	Cooked	
Traditional	260	262	273	263	143	153	1354
Inoculation	323	303	318	312	160	133	1549

Table 4
Comparison of amino acids contents between inoculated and traditional ham (% dry weight)^a

Amino acids	Inoculated	Traditional	Increase
Total quantity	59.46	54.71	8
Cystine	0.80	0.89	-10.11
Tryptophane	0.74	0.64	15.6
Proline	2.12	1.89	12.2
Arginine	3.20	3.41	-6.16
Histidine	2.14	1.92	11.5
Lysine	5.26	4.70	11.9
Phenylalanine	3.11	2.76	12.7
Tyrosine	1.49	1.51	-1.32
Leucine	4.86	4.51	7.8
Isoleucine	3.03	2.76	9.8
Methionine	1.18	1.30	-9.23
Valine	4.16	3.11	33.76
Alanine	3.90	3.21	21.50
Glycine	2.66	2.54	4.70
Glutamate	11.14	9.98	11.6
Serine	2.05	1.99	3.0
Threonine	2.60	2.40	8.30
Aspartate	5.02	5.19	-3.28
Vitamin E (mg/100 g)	47.0	41.0	14.63

^a Sum of soluble and protein amino acids.

underwent a longer curing process, such as Iberian hams, the only taste compounds with a certain importance should be amino acids, peptides and sodium chloride. The protein hydrolysis that took place during the ripening of dry-cured hams was mainly due to endogenous proteolytic activity of cathepsins, calpains and aminopeptidases. The high content of some free amino acids probably contributes to the distinct flavor of Iberian ham. The content of amino acids is related to some odor, too (Martín et al., 2003; Martín, Córdoba, Núñez, Benito, & Asensio, 2004; Ruiz, Ventanas, Cava, Andrés, & García, 1999). One reason of the improved favor of the inoculated Xuanwei ham might be due to the increase of total free amino acid quantity. Amino acids in the hams inoculated by the yeast strain XY8621 and normal ham were analyzed. Comparing to traditional ham, the amino acids of inoculated ham increased 8% (Table 4). This indicated that the yeast inoculated might take part in proteolytic breakdown of protein.

4. Conclusions

There are many kinds of microorganisms growing on the surface of Xuanwei ham. Though moulds have been thought as the contributing microorganism to the quality of Xuanwei ham, the varying growth mode of moulds, the lack of moulds on some high quality hams, and the exist of toxin-producing mould on hams suggested that moulds may not play a good role in ham quality. Therefore, preventing moulds growth was considered beneficial to the procedure of ham process. Yeasts were the predominant fungi on Xuanwei ham. Inoculating yeast on ham at salting time could promote the quality of ham, including the sensory indices of color, flavor, and smell. It also increased the contents of flavor substance such as fragrance and the total amount of free amino acid. Thus, the development of no-mould ham would be beneficial and practicable to the ham industry.

References

- Arnau, J., Gou, P., & Comaposada, J. (2003). Effect of the relative humidity of drying air during the resting period on the composition and appearance of dry-cured ham surface. *Meat Science*, *65*, 1275–1280.
- Brera, C., Miraglia, M., & Colatosti, M. (1998). Evaluation of the impact of mycotoxins on human health: sources of errors. *Microchemical Journal*, *59*, 45–49.
- Creppy, E. E. (2002). Update of survey, regulation and toxic effects of mycotoxins in Europe. *Toxicology Letters*, *127*, 19–28.
- Cvetnić, Z., & Pepeljnjak, S. (1997). Distribution and mycotoxin-producing ability of some fungal isolates from the air. *Atmospheric Environment*, *31*(3), 491–495.
- Deak, T. (1991). Foodborne yeasts. *Advances in Applied Microbiology*, *36*, 179–278.
- De Martinis, E. C. P., & Freitas, F. Z. (2003). Screening of lactic acid bacteria from Brazilian meats for bacteriocin formation. *Food Control*, *14*, 197–200.
- Erdogan, A., Gurses, M., & Sert, S. (2003). Isolation of moulds capable of producing mycotoxins from blue mouldy Tulum cheeses produced in Turkey. *International Journal of Food Microbiology*, *85*, 83–85.
- Escher, F. E., Koehler, P. E., & Ayres, J. C. (1973). Production of ochratoxin A and B on country cured ham. *Applied Microbiology*, *26*, 27–30.
- Fan, X. R., Li, G. W., & Shen, P. (1989). *Test of microbiology* (pp. 260–268) (2nd ed.). Peking, China: High Education Publishing.
- Fields, M. L. (1979). *Mycotoxins of molds. Fundamentals of food microbiology* (pp. 318). Westport Connecticut: AVI Publishing Company Inc.
- GB18357-2001 (2001). *Xuanwei ham*, China.
- Huang, W. K., Zhao, G. J., Lai, X. L., Mao, X. L., & Wei, G. G. (1978). *Food chemical analysis* (p. 28). Shanghai, China: Shanghai Science and Technology Publishing Company.
- Jiang, D. F., Ma, P., Wang, D. Q., Zhang, L. Q., & Jiang, G. Y. (1994). Two new species in *Hansenula* from china ham. *Acta Microbiologica Sinica*, *34*(3), 179–183.
- Kondo, H., & Kato, K. (1983). Plate dilution method. In *The experimental methods of soil organisms* (p. 30). Kaikatado, Tokyo.
- Lin, Z. X. (1998). *Handbook of analysis of ingredient in food*. Peking, China: Light Industry Press of China.
- Losantos, A., Sanabria, C., Cornejo, I., & Carrascosa, A. V. (2000). Characterization of *Enterobacteriaceae* strains isolated from spoiled dry-cured hams. *Food Microbiology*, *17*, 505–512.
- Lücke, F. K. (1986). Microbiological processes in the manufacture of dry sausage and raw ham. *Fleischwirtschaft*, *66*, 1505–1509.
- MacLeod, G. (1996). The scientific and technological basis of meat flavours. In G. G. Birch & M. G. Lindley (Eds.), *Developments in food flavours* (pp. 191–223). London: Elsevier.
- Martín, A., Córdoba, J. J., Benito, M. J., Aranda, E., & Asensio, M. A. (2003). Effect of *Penicillium chrysogenum* and *Debaryomyces hansenii* on the volatile compounds during controlled ripening of pork loins. *International Journal of Food Microbiology*, *84*, 327–338.
- Martín, A., Córdoba, J. J., Núñez, F., Benito, M. J., & Asensio, M. A. (2004). Contribution of a selected fungal population to proteolysis on dry-cured ham. *International Journal of Food Microbiology*, *94*, 55–66.
- Núñez, F., Rodríguez, M. M., Bermúdez, M. E., Córdoba, J. J., & Asensio, M. A. (1996). Composition and toxigenic potential of the mould population on dry-cured Iberian ham. *International Journal of Food Microbiology*, *32*, 185–197.
- Núñez, F., Rodríguez, M. M., Córdoba, J. J., Bermúdez, M. E., & Asensio, M. A. (1996). Yeast population during ripening of dry-cured Iberian ham. *International Journal of Food Microbiology*, *29*, 271–280.
- Portocarrero, S. M., Newman, M., & Mikel, B. (2002). Staphylococcus aureus survival, staphylococcal enterotoxin production and shelf stability of country-cured hams manufactured under different processing procedures. *Meat Science*, *62*, 267–273.
- Rodríguez, M., Núñez, F., Córdoba, J. J., Sanabria, C., Bermúdez, E., & Asensio, M. A. (1994). Characterization of *Staphylococcus* spp. and *Micrococcus* spp. isolated from Iberian ham throughout the ripening process. *International Journal of Food Microbiol*, *24*, 329–335.
- Ruiz, J., Ventanas, J., Cava, R., Andrés, A., & García, C. (1999). Volatile compounds of dry-cured Iberian ham as affected by the length of the curing process. *Meat Science*, *52*, 19–27.
- Saldanha-da-Gama, A., Malfeito-Ferreira, M., & Loureiro, V. (1997). Characterization of yeasts associated with Portuguese pork-based products. *International Journal of Food Microbiology*, *37*, 201–207.

- Stack, M., & Rodricks, J. V. (1971). Methods for analysis and chemical confirmation of sterigmatocystin. *Journal of the Association of Official Agricultural Chemists*, 54, 86–90.
- Su, H. J. J., Chen, H. L., Huang, C. F., Lin, C. Y., Li, F. C., & Donald, K. M. (2002). Airborne fungi and endotoxin concentrations in different areas within textile plants in Taiwan: a 3-year study. *Environmental Research Section A*, 89, 58–65.
- Wu, M. T., Ayres, J. C., & Koehler, P. E. (1974). Production of citrinin by *Penicillium viridicatum* on country-cured ham. *Applied Microbiology*, 27, 427–428.