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The impact of *Lactobacillus brevis* and *Pediococcus pentosaceus* on the sensorial quality of "nem chua" – a Vietnamese fermented meat product

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Abstract: The aim of this study was to investigate the impact of *Lactobacillus brevis* and *Pediococcus pentosaceus* strains isolated from "nem chua", a Vietnamese traditional fermented meat product, on the sensory quality of the product. Three combinations of the two strains (ratios of 1:1, 1:2 and 2:1 respectively) with three levels of bacterial concentration (9.0 x 10^5 CFU/ g, 6.0 x 10^6 CFU/ g and 1.2 x 10^7CFU/ g meat paste) were used as bacterial starters for the fermentation process. The choice of the best starter combination was made by using ranking tests with a jury of 120 Vietnamese tasters. The results showed that the combination of *L. brevis* and *P. pentosaceus* with the total bacterial amount of 6.0 x 10^6 CFU/ g meat paste and the strains ratio of 1:1 conferred the best sensory quality to the fermented products. These findings facilitate further investigations on using and producing these LAB organisms as commercial starters for "nem chua" in Vietnam industry in order to produce products of well-controlled quality and safety.

Key words: Nem chua, *Lactobacillus brevis*, *Pediococcus pentosaceus*, starter, sensory assessment.

Introduction

The addition of competitive starter cultures to begin the fermentation process is an effective method of inhibiting and/or controlling the growth of spoilage organisms, food-borne pathogens and preventing the formation of undesirable end-products. The importance of developing indigenous starters and of increasing knowledge on starters is unwderlined (Talon *et al*., 2007). A preliminary application of microbial starters was introduced in 1950s and since, many studies have discussed their production and their effects on the quality of end products.

In meat product fabrication, the bacterial starters generally used are made-up of a balance between two main groups of bacteria: lactic acid bacteria (*Lactobacillus*) and Gram-positive catalase-positive cocci (*Staphylococcus*) (Hugas *et al*., 1997; Talon *et al*., 2000). Among the *Lactobacillus* species described, *L. plantarum*, *L. brevis, L. curvatus, L. pentosus, L. sakei,* L. *farciminis,* are used frequently. Other bacteria such as various *Pediococcus* species (*P. pentosaceus*, *P. cerevisiae*, *P. acidilactici*), some *Staphylococcus* species (*S. xylosus, S. carnosus, S. simulans*), *Leuconostoc mesenteroides*, and *Lactococcus lactis* are also used in fermented meat products (Montel *et al*., 1996; Coppola *et al*., 1998; Larrouture *et al*., 2000; Erkkilä *et al*., 2001; Ansorena *et al*., 2002; Hughes *et al*., 2002; Paleari *et al*., 2002; Pennacchia *et al*., 2004; Drosinos *et al*., 2005; Klingberg *et al*., 2005; Lee *et al*., 2006 ; Muthukumarasamy *et al*., 2006; Albano *et al*., 2007; Cosansu *et al*., 2007; Edema *et al*., 2007; Latorre-Moratalla *et al*., 2007; Benito *et al*., 2008; Casaburi *et al*., 2008; Hu Yongjin *et al*., 2008; Xiraphi *et al*., 2008; Cenci-Goga *et al*., 2008; Dalmis *et al*., 2008).

"Nem chua" is a very popular fermented meat product in Vietnam. It is made of ground lean pork, spices, sugar, honey, salt, etc, and mixed with boiled pork skin which is cut into thin strings. The meat paste is formed in cubes (2 cm x 3 cm x 3 cm) upon which thin slices of garlic and/or chilli are placed for

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decoration and flavour. The cubes are first wrapped in a piece of leaf that produce a special aroma, then in a plastic film. Banana leaves are usually used as the outer cover. The fermentation lasts about 3 – 4 days at ambient temperature (between 28°C and 30°C). The fermentation takes place either spontaneously, or is orientated by adding a small percentage of fermented meat paste.

In fact, "nem chua" fermentation is unstable and the Vietnamese producers often face problems of quality control due to the proliferation of undesirable microorganisms. The use of imported commercial starters with the intention of preventing the development of harmful flora during natural fermentation does not satisfy the gustative demand of Vietnamese consumers. The development of indigenous starters from lactic acid bacteria isolated from autochthonous products seems to be a reasonable choice.

In the present study, the sensorial effects of culturing *Lactobacillus brevis* and *Pediococcus pentosaceus,* which were selected among lactic acid bacteria identified from "nem chua", was investigated. The aim of this study was to better understand the behaviour of these starters and hopefully to use them for the production of quality-controlled "nem chua" in Vietnam.

Materials and methods

Meat samples

The pork meat paste used to make "nem chua" was supplied by Thu Duc enterprise- a well-known local producer. The meat paste products were produced by the traditional procedure of natural fermentation.

Bacterial starters and cultures

Our preliminary finding indicated that among the five lactic acid bacteria (LAB) species which dominated "nem chua" microbial populations during the five day fermentation, *Lactobacillus brevis* and *Pediococcus pentosaceus* had the highest effect on the sensory quality of the products. Therefore, in this study one strain of *L. brevis* (BE) and one of *P. pentosaceus* (PE) isolated from "nem chua" and well-characterized in our preliminary study (data not shown) were used as starters.

Bacteria in glycerine stocks were refreshed twice in MRS broth (Difco) (24 h, 30^{o}C) and then streaked onto MRS agar (48 h, 30^{o}C). One pure colony was transferred into 10 ml MRS broth (24 h, 30^{o}C). The bacterial cultures were centrifuged (15 min, 2000 x g) and washed twice by centrifugation in MRD solution (Maximum recovery diluent, Difco). Finally, bacterial suspensions were adjusted by dilution with MRD to obtain a density at 600 nm between 0.01 and 0.50 (Hewlett-Packard 8453, USA) and the equivalent bacterial concentration was calculated.

Trial on the impact of each L. brevis *and* P. pentosaceus *strain on the product sensorial quality*

The aim was to determine the appropriate amount of each bacterial strain of *L. brevis* and *P. pentosaceus* to be added to the meat pastes. The experiment was conducted in triplicate as a one-way completely randomized design with three different levels of each bacterial starter: 9.0 x 10^5 CFUs / g meat paste (A), 6.0 x 10^6 CFUs/ g meat paste (B), and 1.2 x 10^7 CFU / g meat paste (C), in which 500g meat paste was used for each time. The pH of each product was monitored during the fermentation using pH meter Testo 230 (Germany). In addition, the sensory quality comparison of products was performed by ranking in hedonic test.

Table 1. Experimental design for the second experiment

Experimental plots	Total numbers of bacterial starters (CFUs/g meat paste)	Ratio of *L. brevis* and *P. pentosaceus*
1 (Aa)	9.0 x 10^5	1:1
2 (Ab)	9.0 x 10^5	1:2
3 (Ac)	9.0 x 10^5	2:1
4 (Ba)	6.0 x 10^6	1:1
5 (Bb)	6.0 x 10^6	1:2
6 (Bc)	6.0 x 10^6	2:1
7 (Ca)	1.2 x 10^7	1:1
8 (Cb)	1.2 x 10^7	1:2
9 (Cc)	1.2 x 10^7	2:1
Control	Starters were not added	Starters were not added

Table 2. Average pH values of "nem chua" meat pastes during the fermention

Compositions	N	$\overline{X}_0 \pm SD$	$\overline{X}_{24} \pm SD$	$\overline{X}_{48} \pm SD$	$\overline{X}_{72} \pm SD$	$\overline{X}_{96} \pm SD$
CONTROL	12	6.02 ± 0.13	5.64 ± 0.26	4.73 ± 0.13	$4.60^{abc} \pm 0.07$	$4.54^{ab} \pm 0.08$
BE_A	5	6.10 ± 0.11	5.70 ± 0.20	4.76 ± 0.23	$4.55^{abcd} \pm 0.11$	$4.47^{abcd} \pm 0.13$
BE_B	10	6.02 ± 0.20	5.57 ± 0.26	4.71 ± 0.18	$4.51^{abcd} \pm 0.11$	$4.39^{abcd} \pm 0.14$
BE_C	5	6.03 ± 0.25	5.30 ± 0.28	4.49 ± 0.16	$4.35^{d} \pm 0.10$	$4.30^{cd} \pm 0.06$
PE_A	5	6.04 ± 0.05	5.55 ± 0.16	4.70 ± 0.07	$4.60^{abc} \pm 0.06$	$4.51^{abc} \pm 0.05$
PE_B	10	6.07 ± 0.11	5.66 ± 0.32	4.83 ± 0.30	$4.61^{abc} \pm 0.07$	$4.53^{ab} \pm 0.05$
PE_C	5	6.00 ± 0.06	5.34 ± 0.20	4.58 ± 0.13	$4.54^{abcd} \pm 0.06$	$4.50^{abcd} \pm 0.03$
PE BE_{Aa}	3	6.17 ± 0.02	5.40 ± 0.11	4.71 ± 0.05	$4.66^{ab} \pm 0.09$	$4.56^{ab} \pm 0.10$
PE BE_{Ab}	3	6.17 ± 0.02	5.42 ± 0.29	4.74 ± 0.10	$4.67^{a} \pm 0.11$	$4.59^{ab} \pm 0.11$
PE BE_{Ac}	3	6.17 ± 0.03	5.53 ± 0.33	4.74 ± 0.10	$4.65^{ab} \pm 0.10$	$4.60^{a} \pm 0.14$
PE BE_{Ba}	3	6.11 ± 0.04	5.27 ± 0.17	4.68 ± 0.06	$4.58^{abc} \pm 0.07$	$4.47^{abcd} \pm 0.08$
PE BE_{Bb}	3	6.16 ± 0.12	5.51 ± 0.27	4.97 ± 0.37	$4.64^{abc} \pm 0.08$	$4.55^{ab} \pm 0.08$
PE BE_{Bc}	3	6.16 ± 0.02	5.40 ± 0.13	4.64 ± 0.04	$4.59^{abc} \pm 0.06$	$4.46^{abcd} \pm 0.06$
PE BE_{Ca}	3	6.14 ± 0.04	5.30 ± 0.12	4.50 ± 0.02	$4.44^{cd} \pm 0.02$	$4.28^{d} \pm 0.07$
PE BE_{Cb}	3	6.14 ± 0.02	5.54 ± 0.39	4.57 ± 0.08	$4.50^{abcd} \pm 0.08$	$4.42^{abcd} \pm 0.07$
PE BE_{Cc}	3	6.15 ± 0.04	5.34 ± 0.12	4.87 ± 0.58	$4.46^{cbd} \pm 0.06$	$4.37^{cbd} \pm 0.06$
P		> 0.05	> 0.05	> 0.05	< 0.001	< 0.001

N = numbers of samples ; $\overline{X}_0$ $\overline{X}_{24}$ $\overline{X}_{48}$ $\overline{X}_{72}$ et $\overline{X}_{96}$: average pH of meat pastes at the beginning, 24 h, 48 h, 72 h và 96 h fermentation, respectively; SD = standard deviation ; P = probability. Mean values followed by different letters within columns are significantly different by Duncan's multiple range tests.

Trial on different combinations of the bacterial starters

Another trial was carried out in triplicate as a two-factor randomized designs experiment. The first factor was the total count of two starters (levels A, B and C as described above) and the other was the ratio of two starters at each level, 1:1 (a), 1:2 (b), and 2:1 (c) proportionally for *L. brevis* and *P. pentosaceus*, respectively (Table 1). The pH of each product was monitored during the fermentation and the sensory quality comparison of end products was carried out by ranking in hedonic test.

To determine the best combination of the bacterial starters

From the above trials three combinations of the bacterial starters of which the fermented products were ranked at more appreciated sensory quality (based on the hedonic assessment described below) were examined again with the control in order to get a composition that conferred the best sensory quality to the products. The experiment was conducted in triplicate as a randomized design with one factor. The pH of each product was monitored during the fermentation. The sensory quality comparison of products was carried out by ranking in hedonic test and the intensity of their sensory attributes was assessed by a descriptive test.

Analysis of organic acids content in the fermented products

Samples of 15 g meat paste were taken during fermentation at 0, 24, 48, 72, and 96 h. Each sample was transferred into a stomacher bag containing 30 mL NaCl 0.85 % and homogenized. The suspension was centrifuged (15 min, 2000 x g) and the upper part was filtrated through a filter membrane with pores of 0.22 µm. Twenty µL of each meat juice were used to determine the amount of organic acids by HPLC (Thermo Separation Products, U.S.) using Aminex HPX-87H column (Bio–Rad laboratory, U.S.). Sulfuric acid 0.01 N was used as diluent with an output of 0.8 µL/ mn at 65°C for 30 mn of detection time.

To determine the preference to the products in hedonic test

The tasting and sensory assessment of the fermented products was carried out at the Sensory Laboratory, Faculty of Food Technologies, Nông Lâm University HCMC. The assessment was performed on the 3rd or 4th day of the fermentation when the products had just ripened ("ripening" products) and obtained the highest quality of flavours (sour and aromatic) and texture. The panel consisted of 60 males and 60 females, 80% of whom were aged between 20 and 30. The geographical origins of the

Table 3. Contents of organic acids produced during "nem chua" fermention in final experiment

		In meat pastes (g/ kg)				
		0h	24h	48h	72h	96h
BE_B	Lactic acid	0	0.38	1.10	4.84	10.47
	Acetic acid	0	0	0	2.08	2.08
	Propionic acid	0	0.02	0.26	0.35	0.52
	Total	0	0.40	1.37	7.27	13.07
PE_B	Lactic acid	0	1.05	1.62	5.13	5.30
	Acetic acid	0	0	0	1.28	1.29
	Propionic acid	0	0.05	0.07	0.08	0.56
	Total	0	1.10	1.69	6.49	7.16
$BEPE_{Bb}$	Lactic acid	0	1.65	4.68	6.92	7.39
	Acetic acid	0	0.32	0.72	0.96	0.91
	Propionic acid	0	0.26	0.23	0.28	0.33
	Total	0	2.23	5.63	8.16	8.63
CONTROL	Lactic acid	0	0.20	2.02	2.87	3.45
	Acetic acid	0	0	0.98	1.14	1.41
	Propionic acid	0	0.05	0.23	0.14	0.07
	Total	0	0.25	3.23	4.15	4.93

Table 4. Average ranks of "nem chua" products analysed with the rangking test

Experiment	Starter combination	n	Σ R	$\overline{R}$ ± SD	$F_{calculated}$	$F_{critical}$		$LSD_{0.05}$
						α=0.05	α=0.01	
First experiment	BE_A	120	281	$2.43^{b} \pm 0.73$	22.50	5.99	9.21	30.36
	BE_B	120	$\underline{229}$	$\underline{1.91}^{a} \pm 0.78$				
	BE_C	120	210	$1.75^{a} \pm 0.83$				
	PE_A	120	260	$2.17^{c} \pm 0.82$	14.60	5.99	9.21	30.36
	PE_B	120	$\underline{206}$	$\underline{1.72}^{d} \pm 0.84$				
	PE_C	120	254	$2.12^{c} \pm 0.71$				
2^e experiment	$PEBE_{Aa}$	60	123	$2.05^{ef} \pm 0.79$	17.45	16.92	21.67	49.58
	$PEBE_{Ab}$	60	124	$2.07^{ef} \pm 0.78$				
	$PEBE_{Ac}$	60	135	$2.25^{ef} \pm 0.80$				
	$PEBE_{Ba}$	60	$\underline{89}$	$\underline{1.48}^{eg} \pm 0.77$				
	$PEBE_{Bb}$	60	139	$2.32^{fh} \pm 0.73$				
	$PEBE_{Bc}$	60	127	$2.12^{ef} \pm 0.74$				
	$PEBE_{Ca}$	60	94	$1.57^{ef} \pm 0.67$				
	$PEBE_{Cb}$	60	122	$2.03^{ef} \pm 0.86$				
	$PEBE_{Cc}$	60	123	$2.05^{ef} \pm 0.79$				
	CONTROL	60	124	$2.07^{ef} \pm 0.88$				
Final experiment	BE_B	120	271	$2.26^{i} \pm 1.05$	31.78	7.81	11.34	33.95
	PE_B	120	305	$2.54^{j} \pm 1.03$				
	$PEBE_{Ba}$	120	261	$\underline{2.18}^{i} \pm 1.18$				
	CONTROL	120	363	$3.03^{k} \pm 1.03$				

n = total taster number ; Σ R = total rangs ; $\overline{R}$ = average of rangs.
Mean values followed by different letters within columns are significantly different by LSD test.

panel were representative. Samples were evalued using a "liking level" category scale (5 levels) and ranked on a hedonic tasted preference scale. The test products were randomly coded and presented in similar appearance of 10 g cubes and served at room temperature (20-25 °C). The samples were presented in random positions for tasting but the overall numbers of each tasting position for each product were equal. In second experiment, the display positions of each product were arranged using a balanced incomplete block design (BIBD) for three of the ten samples in all. In a single pass through the BIBD, each sample had evaluated three of the ten assessors and each pair of samples was evaluated by two assessors. The entire BIBD was repeated twenty times to acquire finally sixty evaluations for each product.

Assessment of the intensities of "nem chua" sensory attributes by descriptive test

The establishment of the "nem chua" sensory profile was based on the standard ISO 11035:1994 – Guidance for identification and selection of descriptors by a multidimensional approach. The colour, brightness of surface, firmness, cohesiveness, sourness and sour aroma, leafy aroma, sweetness, bitterness, alcoholic flavour, alcoholic odour, garlic flavour, garlic odour, saltiness, and umaminess were the 15 assessed sensory attributes. A panel of 8 members was established. These members were students who had taken the course of sensory assessment. The members were trained for 30 h for the assessment of the sensorial attributes of "nem chua" and reached agreement on a 10-point universal intensity scale. The training allowed the panellists to acquire a consensus assessment of the sensory attributes and assure the reproductivity of this assessment (followed the standard ISO 8586–1:1993).

Statistical analysis

In the ranking tests, the data were analyzed using Friedman's test and the significance of difference was calculated by LSD (Least Significant Difference, ISO 8587:2006). The analysis of pH data and the assessment of the intensities were performed by Fisher test. The Principal Components Analysis was used to determine their relationships and the Partial Least Squares Regression (PLSR) to understand the relation between the descriptive data and the preference scores of consumers. All analyses were performed by MINITAB 15.

Results

pH changes in the meat paste during fermentation

The pH values of the experimental meat pastes measured at different stages of the fermentation are presented in Table 2. The results indicated significant pH reduction of the products ($p<0.001$), which progressively decreased during the fermentation process. During the first 24 hours of fermentation, the pH of meat pastes supplemented with LAB decreased by 0.40 - 1.04 compared to 0.39 of the control group. The pH of the products cultured with both *L. brevis* and *P. pentosaceus* strains decreased significantly more than that in the products with a single or in the control group. However, from 24 h to 48 h of fermentation the pH of the latter decreased more quickly than that of the former. After two days fermentation, only a slight reduction of the pH in all products was observed. The change of pH values in every fermented product was not significant between 72 h and 96 h of the fermentation ($p > 0.05$).

The difference became significant after 72 h fermentation ($p<0.001$). At 96 h of fermentation, the products inoculated with *L. brevis* or their combinations of 1.2 x 10^7 CFU/g meat paste had the lowest pH (4.34 ± 0.07). The pH of the others was approximately 4.50.

The analyses of the contents of different organic acids in all products in experiment 3 showed that the acidity of the fermented meats was mainly due to lactic acid (Table 3). At the end of the fermentation process, among LAB-inoculated products the highest levels of lactic acid were detected in those inoculated with *L. brevis* (10.47 g lactic acids/ kg meat paste; accounting for 80.14% of the total acids produced) and the lowest lactic acid contents were found from those cultured with *P. pentosaceus* (5.30 g/ kg meat paste, accounting for 74.11% of the total acids produced). The control products comprised of 3.45 g lactic acid per kilogram meat paste (70.02% of the total acids produced). Acetic acid was detected from all fermented products at low levels (ranging from 1.29 to 2.08g/ kg meat paste, making up 15.90 - 17.46% of the total acids produced).

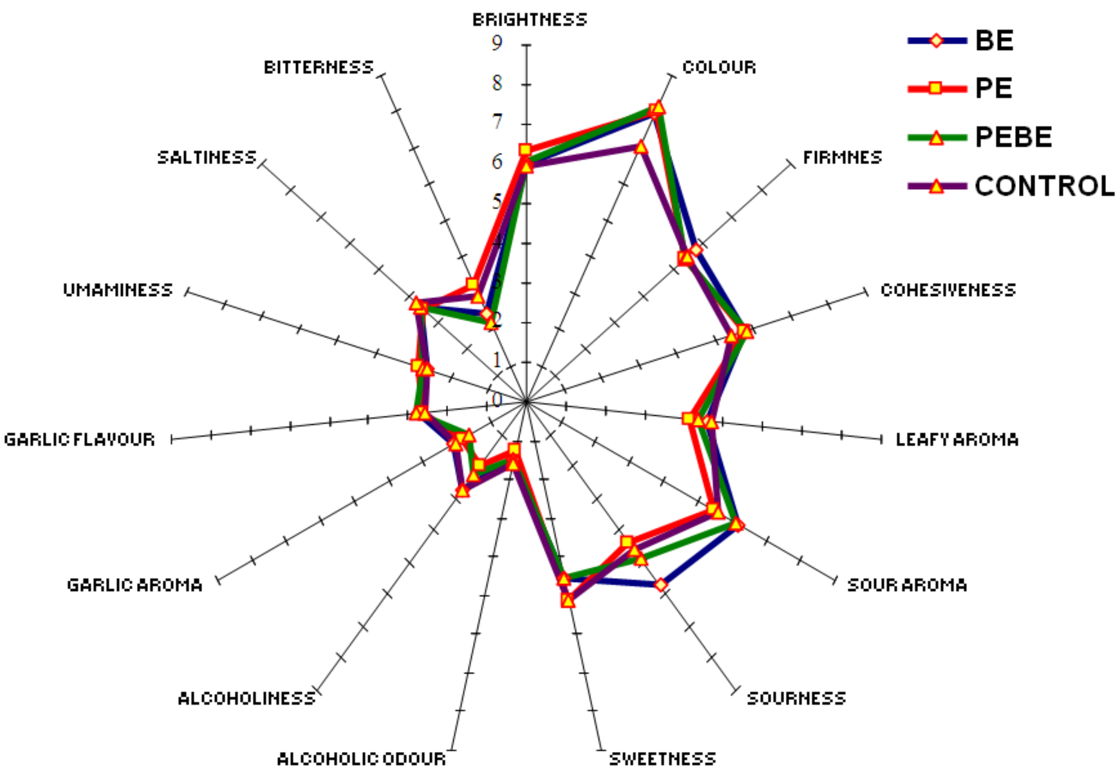

Figure 1. The intensities of sensorial attributes of "nem chua" products of the final experiment

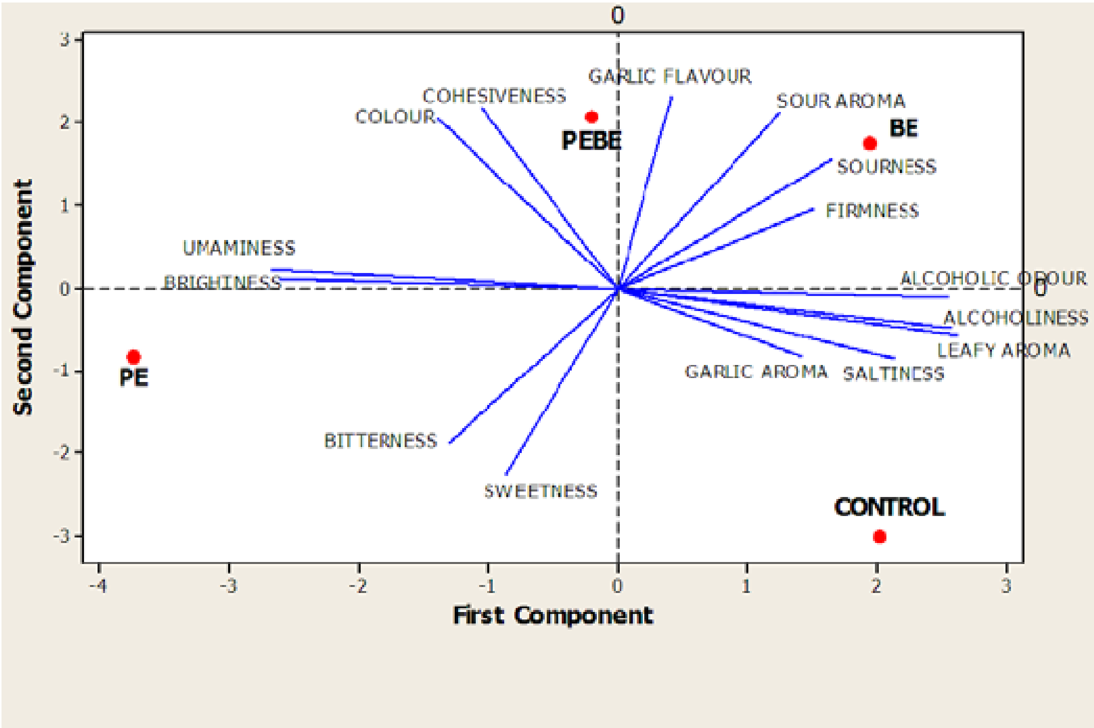

Figure 2. Correlation among the evaluated sensorial attributes of "nem chua" by Principal Components Analysis

Sensory assessment of the products

The results of the hedonic assessment of fermented products are given in Table 4. A significant difference ($p<0.01$) in consumer preference was found among fermented products in first experiment, which indicated the effects of bacterial concentration in the starter (of either *L. brevis* or *P. pentosaceus*) on the sensory of the respective products ($F_{calculated}$ for *L. brevis* and *P. pentosaceus* was 22.5 and 14.6, respectively, $F_{critical}$ = 9.21 at a significance level of 99%). Among the products cultured with *P. pentosaceus*, those inoculated with 6.0 x 10^6 CFU/ g meat paste (level B) obtained the highest ranks of the assessment. For *L. brevis,* similarly high quality was recorded for both products inoculated with 6 x 10^6 CFU bacteria per g meat paste (level B) and 1.2 x 10^7 CFU per gram (level C) ($p > 0.05$).

In second experiment, different combinations of the bacterial starters were seen to significantly influence the product quality ($F_{calculated}$= 17.45 > $F_{critical}$ = 16.92 at a significance level of 95%). Similar results were obtained for all experimental combinations with respect to bacterial strains and their counts and ratios in the starters. The best quality assessment was given to the products of combination $PEBE_{Ba}$ (total inocula of 6.0 x 10^6 CFU/g meat paste, the ratio of *L. brevis* and *P. pentosaceus* was 1:1).

Similarly, the assessment in final experiment indicated significant impacts of bacterial combinations in the starters to fermented products ($p < 0.01$). The products of composition $PEBE_{Ba}$ were still given the best quality. However, there was no statistical difference in the sensory assessment between these products and those inoculated with only *L. brevis* ($p > 0.05$).

For our study, the "nem chua"s supplemented with both *L. brevis* and *P. pentosaceus* strains (total inocula of 6.0 x 10^6 CFU/ g meat paste and strains ratio of 1:1) were the most preferred owing to their sweet- and sour- flavour harmony and the attractive pink colour. The control products and those added with *P. pentosaceus* (PE) possessed a more bitter taste compared to others. The control products without starter addition were least preferred due to their pale colour, and less flavour harmony (less sour and more salty and sweet). In addition, the results of the microbiological analyses (made by the service of sciences, technologies and environment of Ho Chi Minh City), showed that the microbiological quality of the products with starters was higher than the control (data not shown).

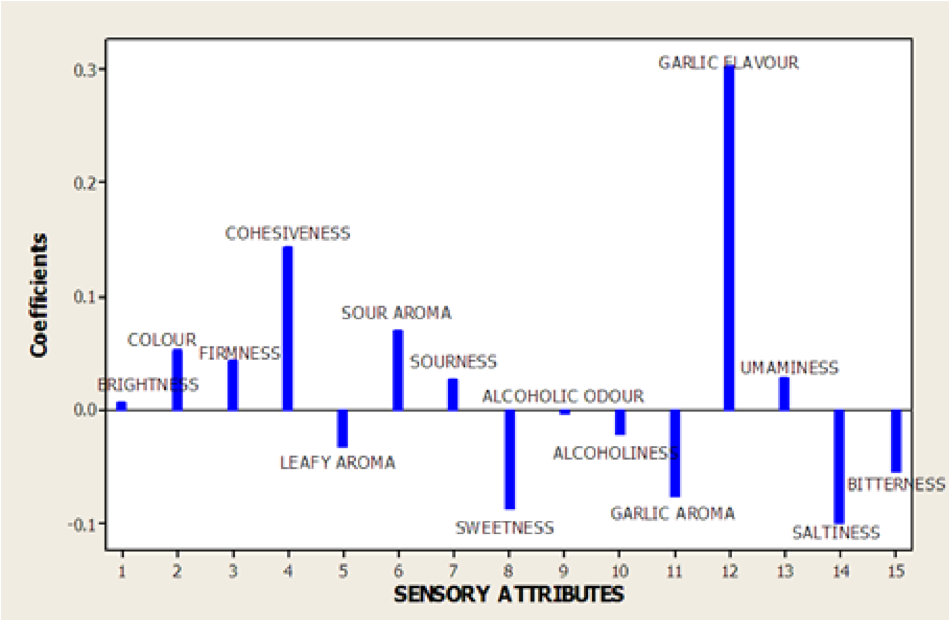

Figure 3. Correlation between Nem chua sensorial attributes and the Vietnamese preference (by PLS Coefficent Plot)

Assessment of the intensity of nem chua sensory attributes

The results of the product assessment in final experiment are presented in Figure 1. The statistical analyses showed significant differences ($p < 0.05$) in the colour, firmness, and cohesiveness of the product texture, sourness and sour aroma, leafy aroma, sweetness, bitterness and alcoholic flavour of the products. No statistical difference was noted in the brightness of surface, alcoholic odour, garlic flavour, saltiness, and umaminess between these products.

The PCA separated four nem chua products in two groups: group 1 included BE and PEBE; and group 2 consisted of PE and the control (Figure 2, with PCA1 explaining 48.5% and PCA2 explaining 38.2% of variance). Products of group 1 were characterized by sour flavour, firmness, and cohesiveness of texture, and pink turn colour. Group 2 presented alcoholic flavour, leafy aroma, saltiness, and sweetness.

The PLS analysis (Figure 3) showed significant correlations ($p<0.05$) between the preference of Vietnamese consumers with the product sweetness (-0.921), colour (0.883), sour aroma (0.852), cohesiveness (0.927), garlic flavour (0.971), and bitterness (-0.779). The correlation was positive for the surface brightness, colour, texture firmness and cohesiveness, sour flavour and umaminess. It was negative for the leafy aroma, alcoholic and garlic flavour, bitterness, sweetness and saltiness. Because of the small size of samples, it should be noted that the study correlations should be considered as an exploratory analysis facilitating the assessment of the sensorial attributes from the preferences on the products.

Discussion

In meat fermentation, the main function of LAB is to obtain a rapid pH drop of the paste, which in turn increases product stability and shelf-life by inhibiting undesirable spoilage microorganisms. The LAB also create biochemical conditions to attain the new sensory properties of the ripe products through modification of the raw materials (Hammes and Hertel, 1998; Lücke, 2000 cited by Ammor *et al*., 2007). In the traditionally fermented sausage fabrication, the great diversity of lactic acid bacteria and staphylococci was linked to manufacturing practices. The development of indigenous starters is very promising because it enables sausages to be produced with both high sanitary and sensory qualities (Talon *et al*., 2007).

In the present study, it was shown that the fermentation with *L. brevis* and/or *P. pentosaceus* as starters produced "nem chua" of lower pH compared to the natural process without bacterial addition. In addition, it took three days for the former to reach pH of 4.5 while it was four days for the latter. The lower pH in our products was obtained with the higher concentrations of starter bacteria, especially when *L. brevis* was used in the starter combination. The molar ratio of lactic acid / acetic acid (L/A) was greated than 1. However, L/A ratio much less than 1 (1 :7) were noted by Lee *et al.* (2006) for the same bacteria, but in a fluid model medium modified according to the special conditions of fermented sausages. The differences in the ratio L/A are certainly due to the differences in the matrix.

In our study, the products obtained with *L. brevis* starters had a highly pronounced acid taste. This is related to the high total quantity of acids produced. The quantity of acids produced diminishes when *P. pentosaceus* are added to *L. brevis* or when *P. pentosaceus* alone are used as a starter. In this latter case, the acid taste is not sufficiently pronounced. The product preferred by the jury was obtained by a combination of the two strains used as a starter.

González-Fernández *et al*. (2006) studied the instrumental and sensory textural properties of chorizo with different starter cultures. In general terms, changes of pH over time are influenced by the presence of starter culture even if all chorizos showed at first a rapid decrease, a stay or slow decrease, and a final rise. As with the results for the "Nem chua", the control chorizo sausages showed a slower pH drop than the chorizo sausages inoculated with starter cultures. González-Fernández et al. showed that the pH diminution was much quicker with *Lactobacillus* (*L. sakei* K29) than with *Pediococcus*. The firmness, hardness and chewiness tended to increase with time in all sausages and reached the highest values in chorizo with *Lactobacillus*, followed by values in chorizo with *Pediococcus*. The lowest values were in chorizo without starter culture. We noted equally that the best observed Nem chua texture results occurred when *Lactobacillus* were present in the starter (either alone or in combination with *Pediococcus*). The major changes in structure take place in the fermentation process would be suggested by the pH reduction, which induces the aggregation of proteins leading to the formation of an ordered protein network, contributing to firmness.

For both the acid taste and the texture of Nem chua, the best results were obtained by a combination of the two strains (*L. brevis* and *P. pentosaceus*). The role of *P. pentosaceus* is also important for another reason. The study of Hu *et al*. (2008) showed that *P. pentosaceus* produced antioxidant effects on unsaturated fatty acids. Thiobarbituric acid (TBA) value of products added with this bacterium was definitely lower than that found in the control. This remark confirmed the result from Tjener *et al*. (2003) and Talon *et al*. (2000) about the antioxidant action of starter *P. pentosaceus* in lipid oxidation. It would be interesting to study the similar effect of *Pediococcus pentosaceus* in the product "Nem chua".

Conclusion

In this report we are determined the nature of starter and concentrations which produce a Nem chua whose taste, texture, colour and flavour correspond to typical Vietnamese tastes. Namely a combination of *L. brevis* and *P. pentosaceus* with the total bacterial amount of 6.0 x 10^6 CFU/ g meat paste and the strains ratio of 1:1. The experiments show that the sensorial quality obtained is reproducible. This latter point is essential in the quality control of an industrial production.

References

Albano, H., Oliveira, M., Aroso, R., Cubero, N., Hogg, T. and Teixeira, P. 2007. Antilisterial activity of lactic acid bacteria isolated from "Alheiras" (traditional Portuguese fermented sausages) : In situ assays. Meat Science 76 (4): 796-800.

Ammor, M. S. and Mayo, B. 2007. Selection criteria for lactic acid bacteria to be used as functional starter cultures in dry sausage production: An update. Meat Science 76 (1): 138-146.

Ansorena, D., Montel, M. C., Rokka, M., Talon, R., Eerola, S., Rizzo, A., Raemaekers, M., and Demeyer, D. 2002. Analysis of biogenic amines in northern and southern European sausages and role of flora in amine production. Meat Science 61 (2): 141-147.

Benito, M.J., Serradilla, M.J., Ruiz-Moyano, S.M., Alberto P.-N., and Francisco Córdoba, M.G. 2008. Rapid differentiation of lactic acid bacteria from autochthonous fermentation of Iberian dry-fermented sausages. Meat Science In Press. Corrected Proof.

Casaburi, A., Di Monaco, R., Cavella, S., Toldrá, F., Ercolini, D., and Villani, F. 2008. Proteolytic and lipolytic starter cultures and their effect on traditional fermented sausages ripening and sensory traits. Food Microbiology 25 (2): 335-347.

Cenci-Goga, B.T., Ranucci, D., Miraglia, D., and Cioffi, A. 2008. Use of starter cultures of dairy origin in the production of Salame Nostrano, an Italian dry-cured sausage. Meat Science 78 (4): 381-390.

Coppola, R., Giagnacovo, B., Iorizzo, and M., Grazia, L. 1998. Characterization of lactobacilli involved in the ripening of Soppressata Molisana, a typical southern Italy fermented sausage. Food Microbiology 15 (3): 347-353.

Cosansu, S., Kuleasan H., Ayhan, K., and Materon, L. 2007. Antimicrobial activity and protein profiles of *Pediococcus* spp. isolated from Turkish "Sucuk". Journal of Food Processing and Preservation 31 (2): 190-200.

Dalmis, Ü. and Soyer, A. 2008. Effect of processing methods and starter culture (*Staphylococcus xylosus* and *Pediococcus pentosaceus*) on proteolytic changes in Turkish sausages (Sucuk) during ripening and storage. Meat Science In Press. Corrected Proof.

Drosinos, E.H., Mataragas, M., Xiraphi, N., Moschonas, G., Gaitis, F., and Metaxopoulos, J. 2005. Characterization of the microbial flora from a traditional Greek fermented sausage. Meat Science 69 (2): 307-317.

Edema, M. O. and Sanni, A. I. 2007. Functional properties of selected starter cultures for sour maize bread. Food Microbiology 25 (4): 616-625.

Erkkilä, S., Petäjä, E., Eerola, S., Lilleberg, L., Mattila-Sandholm, and T., Suihko, M. 2001. Flavour profiles of dry sausages fermented by selected novel meat starter cultures. Meat Science 58 (2): 111-116.

González-Fernández, C., Santos, E.M., Rovira, J., and Jaime, I. 2006. The effect of sugar concentration and starter culture on instrumental and sensory textural properties of chorizo-Spanish dry-cured sausage. Meat Science 74 (3): 467-475.

Hammes, W. P. and Hertel, C. 1998. New developments in meat starter cultures. Meat Science. Meat Consumption and Culture 44th International Congress of Meat Science and Technology 49 (Supplement 1): S125-S138.

Hu, Yongjin, Xia, Wenshui and Ge, Changrong. 2008. Characterization of fermented silver carp sausages inoculated with mixed starter culture. LWT - Food Science and Technology 41 (4): 730-738.

Hugas, M. and Monfort J. M. 1997. Bacterial starter cultures for meat fermentation. Food Chemistry 59 (4): 547-554.

Hughes, M. C., Kerry, J. P., Arendt, E. K., Kenneally, P. M., McSweeney, P. L. H., and O'Neill, E. E. 2002. Characterization of proteolysis during the ripening of semi-dry fermented sausages. Meat Science 62 (2): 205-216.

Klingberg, T.D., Axelsson, L., Naterstad, K., Elsser, D., and Budde, B. B. 2005. Identification of potential probiotic starter cultures for Scandinavian-type fermented sausages. International Journal of Food Microbiology 105 (3): 419-431.

Larrouture, C., Ardaillon, V., Pépin, M., and Montel, M. C. 2000. Ability of meat starter cultures to catabolize leucine and evaluation of the degradation products by using an HPLC method. Food Microbiology 17 (5): 563-570.

Latorre-Moratalla, M.L., Bover-Cid, S., Aymerich, T., Marcos, B., Vidal-Carou, M.C., and Garriga, M. 2007. Aminogenesis control in fermented sausages manufactured with pressurized meat batter and starter culture. Meat Science 75 (3): 460-469.

Lee, Joo-Yeon, Kim, Cheon-Jei and Kunz, BennoLee. 2006. Identification of lactic acid bacteria isolated from kimchi and studies on their suitability for application as starter culture in the production of fermented sausages. Meat Science 72 (3): 437-445.

Paleari M.A., Bersani C., Vittorio M.M., and Beretta G. 2002. Effect of curing and fermentation on the microflora of meat of various animal species. Food Control 13: 195–197.

Montel, M. -C., Reitz, J., Talon, R., Berdagué, J. -L., and Rousset-Akrim, S. 1996. Biochemical activities of *Micrococcaceae* and their effects on the aromatic profiles and odours of a dry sausage model. Food Microbiology 13 (6): 489-499.

Muthukumarasamy, P. and Holley, R. A. 2006. Microbiological and sensory quality of dry fermented sausages containing alginate-microencapsulated *Lactobacillus reuteri*. International Journal of Food Microbiology 111 (2): 164-169.

Pennacchia, C., Vaughan, E.E. and Villani, F. 2006. Potential probiotic *Lactobacillus* strains from fermented sausages: Further investigations on their probiotic properties. Meat Science 73 (1): 90-101.

Standard ISO 8587:2006 – Sensory analysis – Methodology – Rangking

Standard ISO 11035:1994 – Guidance for identification and selection of descriptors by a multidimensional approach.

Standard ISO 8586 – 1 : 1993 – Sensory analysis – General guidance for the selection, training and monitoring of assessor.

Talon, R., Walter, D. and Montel, M. C. 2000. Growth and effect of staphylococci and lactic acid bacteria on unsaturated free fatty acids. Meat Science 54 (1): 41-47.

Talon, R., Leroy, S. and Lebert, I. 2007. Microbial ecosystems of traditional fermented meat products: The importance of indigenous starters. Meat Science 77 (1): 55-62.

Tjener, K., Stahnke, Louise H., Andersen, L., and Martinussen, J. 2004. A fermented meat model system for studies of microbial aroma formation. Meat Science 66 (1): 211-218.

Xiraphi, N., Georgalaki, M., Rantsiou, K., Cocolin, L., Tsakalidou, and E., Drosinos, E.H. 2008. Purification and characterization of a bacteriocin produced by *Leuconostoc mesenteroides* E131. Meat Science In Press. Corrected Proof.