ACTA SCIENTIARUM POLONORUM
Acta Sci. Pol. Technol. Aliment. 14(4) 2015, 407–414
www.food.actapol.net pISSN 1644-0730 eISSN 1889-9594 DOI: 10.17306/J.AFS.2015.4.40

# EFFECT OF BACTERIA PROPORTION ON THE FERMENTATION OF GOAT YOGHURT WITH PROBIOTIC CULTURE

Guowei Shu[1]✉, Shuai Wang[1], Zikun Chen[2], He Chen[1], Changfeng Wang[1], Yaning Ma[3]

[1]School of Food and Biological Engineering, Shaanxi University of Science and Technology
Xi'an, 710021, **China**
[2]Xinxiang Institute for Food and Drug Control, Henan Province
Xinxiang, 453000, **China**
[3]Shangqiu Entry-Exit Inspection and Quarantine Bureau, Henan Province
Shangqiu, 476000, **China**

## ABSTRACT

**Background.** Goat milk production in Shaanxi province is dominant in China, but the product is mainly infant formula and adult milk powder; product homogeneity is serious and has no goat yoghurt with probiotic culture.
**Methods.** The effect of bacteria proportion (1:3:1, 1:2:1, 1:1:1, 2:1:1, 3:1:1) on pH, acidity, and viable counts and sensory evaluation of goat milk fermented by probiotics including *L. acidophilus*, *B. bifidum* or *L. casei besides*, *S. thermophilus* and *L. bulgaricus* for developing AB-goat yoghurt and BC-goat yoghurt was investigated.
**Results.** The optimum bacteria proportion of *L. acidophilus* : *B. bifidum* : *S. thermophilus* and *L. bulgaricus* for AB-goat yoghurt and *B. bifidum* : *L. casei* : *S. thermophilus* and *L. bulgaricus* for BC-goat yoghurt were both 2:1:1. The pH, acidity, the viable counts of *L. acidophilus* and *B. bifidum*, the total viable counts were respectively 4.60, 7.73 (g/L), $3.50\times10^7$ cfu/mL, $3.40\times10^7$ cfu/mL and $2.30\times10^9$ cfu/mL in AB-goat yoghurt. The pH, acidity, the viable counts of *B. bifidum* and *L. casei*, the total viable counts were respectively 4.61, 8.16 (g/L), $7.60\times10^7$ cfu/mL, $5.60\times10^7$ cfu/mL and $2.04\times10^9$ cfu/mL in BC-goat yoghurt.
**Conclusion.** The bacteria proportion had a significant effect on fermentation of AB- and BC-goat yoghurt, the results are beneficial for developing AB-goat yoghurt and BC-goat yoghurt.

**Key words:** goat milk, bacteria proportion, *L. acidophilus*, *B. bifidum*, *L. casei*, yoghurt

## INTRODUCTION

Milk products from goat, such as yoghurt and cheese, are becoming increasingly popular in developed countries as substitutes for cow milk products for those who have allergies to cow milk (Haenlein, 2004; Park, 1994). Goat milk production in Shaanxi province amounts to a total of 650,000 tons in 2014, but the product is mainly infant formula and adult milk powder; product homogeneity is serious and has no goat yoghurt.

Yoghurt that contains probiotics such as *L. acidophilus*, *B. bifidum* and *L. casei* is also becoming popular because probiotics can provide various health benefits such as prevention of gastrointestinal disorders, diarrhea and colon cancer, and alleviating lactose intolerance and improving immunity (Kailasapathy and Chin, 2000; Krasaekoopt et al., 2003; Rolfe, 2000; Singh et al., 2011). Posecion et al. (2005) established

✉shuguowei@gmail.com

process conditions suitable for a small-scale goat milk yoghurt and examined the physicochemical properties and organoleptic acceptability of the resultant product. Drinkable yoghurts made with normal and probiotic cultures were evaluated for their sensory characteristics (Uysal-Pala et al., 2006). Quality characteristics of yoghurt from goat milk supplemented with aronia juice and blueberry juice was investigated (Boycheva et al., 2011). Mutlu and Akin (2007) researched effects of cysteine and different incubation temperatures on the microflora, chemical composition and sensory characteristics of bio-yoghurt made from goat's milk. Farnsworth et al. (2006) investigated the effects of transglutaminase treatment on functional properties and probiotic culture survivability of goat milk yoghurt, but the bacteria proportion on goat milk fermented by probiotic culture was not studied.

In our previous study, the process of fermentation set-style type goat yoghurts was optimized by *S. thermophilus* and *L. bulgaricus* (Chen et al., 2010), the effect of inoculum and temperature on the fermentation of goat yoghurt by *L. bulgaricus* and *S. thermophilus* (Shu et al., 2014) was investigated, the effect of the total inoculum size containing *L. acidophilus* or *L. casei* on the fermentation of goat milk was studied on the basis of *S. thermophilus* and *L. bulgaricus* as starter cultures (Chen et al., 2015). The purpose of the present work was to study the effect of bacteria proportion on goat milk fermented by probiotics including *L. acidophilus*, *B. bifidum* or *L. casei* besides, *S. thermophilus* and *L. bulgaricus* for developing goat yoghurt containing *L. acidophilus* and *B. bifidum* (AB-goat yoghurt) and goat yoghurt containing *B. bifidum* and *L. casei* (BC-goat yoghurt).

## MATERIAL AND METHOD

### Materials and reagents

Fresh goat milk mainly composed of fat (3.75 g/100 g), lactose (4.13 g/100 g), protein (3.42 g/100 g), ash (0.78 g/100 g) and total solids (12.08 g/100 g) was purchased from local farmers (Xi'an Weiyang, China). All chemicals used were of analytical grade unless otherwise specified.

### Microorganism

*L. acidophilus* (LA), *Bifidobacterium bifidum* (BB), *L. casei* (LC), *S. thermophilus* (ST) and *L. bulgaricus* (LB) were obtained from School of Food and Biological Engineering, Shaanxi University of Science and Technology. They were inoculated three successive times with MRS (for *L. acidophilus*, *B. bifidum*, *L. casei* and *L. bulgaricus*) and M17 (for *S. thermophilus*) broth (Hopebio, Qindao, China) in order to obtain fresh culture. The activated LA, BB or LC was inoculated into sterilized goat milk at 5% inoculum size, mixed and cultivated at 37°C until coagulation, respectively. The activated *S. thermophilus* and *L. bulgaricus* were inoculated into sterilized goat milk at 5% inoculum size with 1 : 1 ratio, mixed and cultivated in the incubators until coagulation; they would be used for the production of AB or BC-goat yoghurt.

### Preparation of probiotic goat yoghurts

Goat milk was heated to 90°C for 10 minutes, cooled to 45°C, and then was divided into equal portions in order to prepare AB-goat yoghurt BC-goat yoghurt, the bacteria proportion (v/v) of *L. acidophilus*, *B. bifidum*, *S. thermophilus* and *L. bulgaricus* for AB-goat yoghurt were 1:3:1, 1:2:1, 1:1:1, 2:1:1, 3:1:1, the bacteria proportion (v/v) of *B. bifidum*, *L. casei*, *S. thermophilus* and *L. bulgaricus* BC-goat yoghurt was the same as AB-goat yoghurt. The mixed liquid starter cultures of different bacteria proportion were inoculated into goat milk at 5% total inoculum size mentioned above, then fermented at 39°C constant temperature. The acidity, pH value, viable counts of *L. acidophilus*, *B. bifidum* or *L. casei*, total viable bacteria were determined every other 1.5 h, then given a sensory evaluation after 12 h in cold storage so as to study the influence of bacteria proportion on the fermentation of goat yoghurt. All samples were carried out by triplicate, each sample was measured three times and the results are the average of three samples.

### Analysis methods

Plate coating method was used to determinate the viable counts, the total viable counts were determinate by modified Tomato Juice medium, determination of *L. acidophilus* or *L. casein* by MRS agar containing 0.06% bile salt and determination of *B. bifidum* by MRS agar containing 0.10% LiCl (Chen et al., 2011; Shu et al., 2011). The pH of culture media was directly determined through a pH-meter (pHS-3c) at the room temperature, acidity was determined by titration with

0.1N NaOH and expressed in lactic acid content (g/L). The samples were organoleptically assessed by five panelists, tasted and described the texture of the product: colour, texture, taste, smell, who were trained on the basis of normal sensory acuity and consistency.

## RESULTS AND DISCUSSION

### Effect of bacteria proportion on the fermentation of AB-goat yoghurt

The effect of bacteria proportion on the fermentation of AB-goat yoghurt was shown in Figure 1 and Table 1.

Figure 1a showed viable counts of *L. acidophilus* in all AB-goat yoghurt samples first increased within 4.5 h, and then decreased between 4.5 h and 6.0 h at different bacteria proportion. The viable counts of *L. acidophilus* first increased from $1.83\times10^7$ cfu/ml at 1.5 h to $3.46\times10^7$ cfu/ml at 4.5 h, and then decreased to $2.70\times10^7$ cfu/ml at 6 h when the bacteria proportion was 1:3:1. The viable counts of *L. acidophilus* first increased from $1.50\times10^7$ cfu/ml at 1.5 h to $3.40\times10^7$ cfu/ml at 4.5 h, and then decreased to $2.84\times10^7$ cfu/ml at 6 h when the bacteria proportion was 1:2:1. The viable counts of *L. acidophilus* first increased from $1.33\times10^7$ cfu/ml at 1.5 h to $3.39\times10^7$ cfu/ml at 4.5 h, and then decreased to $2.32\times10^7$ cfu/ml at 6 h when the bacteria proportion was 1:1:1. The viable counts of *L. acidophilus* first increased from $1.43\times10^7$ cfu/ml at 1.5 h to $3.50\times10^7$ cfu/ml at 4.5 h, and then decreased to $2.70\times10^7$ cfu/ml at 6 h when the bacteria proportion was 2:1:1. The viable counts of *L. acidophilus* first increased from $1.07\times10^7$ cfu/ml at 1.5 h to $3.80\times10^7$ cfu/ml at 4.5 h, and then decreased to $3.60\times10^7$ cfu/ml at 6 h when the bacteria proportion was 3:1:1. Among them, the viable counts of *L. acidophilus* at 3:1:1 bacteria proportion reached the maximum ($3.60\times10^7$ cfu/mL), the viable counts at 1:1:1 bacteria proportion was the lowest ($3.39\times10^7$ cfu/ml) at 4.5 h.

Figure 1b showed the change of viable counts of *B. bifidum* in all AB-goat yoghurt samples was the same as *L. acidophilus*. The viable counts of *B. bifidum* first increased quickly within 3 h, then increased slowly to maximum between 3.0 and 4.5 h, then decreased between 4.5 and 6.0 h. Among them, the viable counts of *B. bifidum* at 2:1:1 bacteria proportion reached the maximum ($3.40\times10^7$ cfu/mL), the viable counts of *B. bifidum* at 1:3:1 bacteria proportion was the lowest ($2.11\times10^7$ cfu/ml).

Figure 1c showed that the total viable counts in AB-goat yoghurt were all first increased then decreased as *L. acidophilus* or *B. bifidum* with time increasing. Among them, the total viable counts at 3:1:1 bacteria proportion reached the maximum ($2.50\times10^9$ cfu/mL) followed at 3:1:1 ($2.30\times10^9$ cfu/mL), the total viable counts at 1:1:1 bacteria proportion was the minimum ($1.79\times10^9$ cfu/mL). From Figure 1d and 1e, pH in all AB-goat yoghurt decreased rapidly within 4.5 h, then began to slow down, while the variation trend of acidity in all AB-goat yoghurt was opposite to pH. The acidity and pH at 3:1:1, 2:1:1 bacteria proportion was 7.84 (g/L), 7.73 (g/L) and 4.59, 4.60 for 4.5 h, respectively.

**Table 1.** The sensory evaluation of AB-goat yoghurt fermented by different bacteria proportion

| Bacteria proportion of LA:BB:LB+ST | Colour | Smell | Taste | Texture | Comprehensive evaluation |
|---|---|---|---|---|---|
| 1:3:1 | 0.8 | 1.76 | 1.89 | 2.36 | 6.81 |
| 1:2:1 | 0.8 | 1.72 | 2.00 | 2.39 | 6.91 |
| 1:1:1 | 0.8 | 1.84 | 2.03 | 2.39 | 7.06 |
| 2:1:1 | 0.8 | 1.86 | 2.24 | 2.40 | 7.30 |
| 3:1:1 | 0.8 | 1.76 | 2.00 | 2.37 | 6.93 |

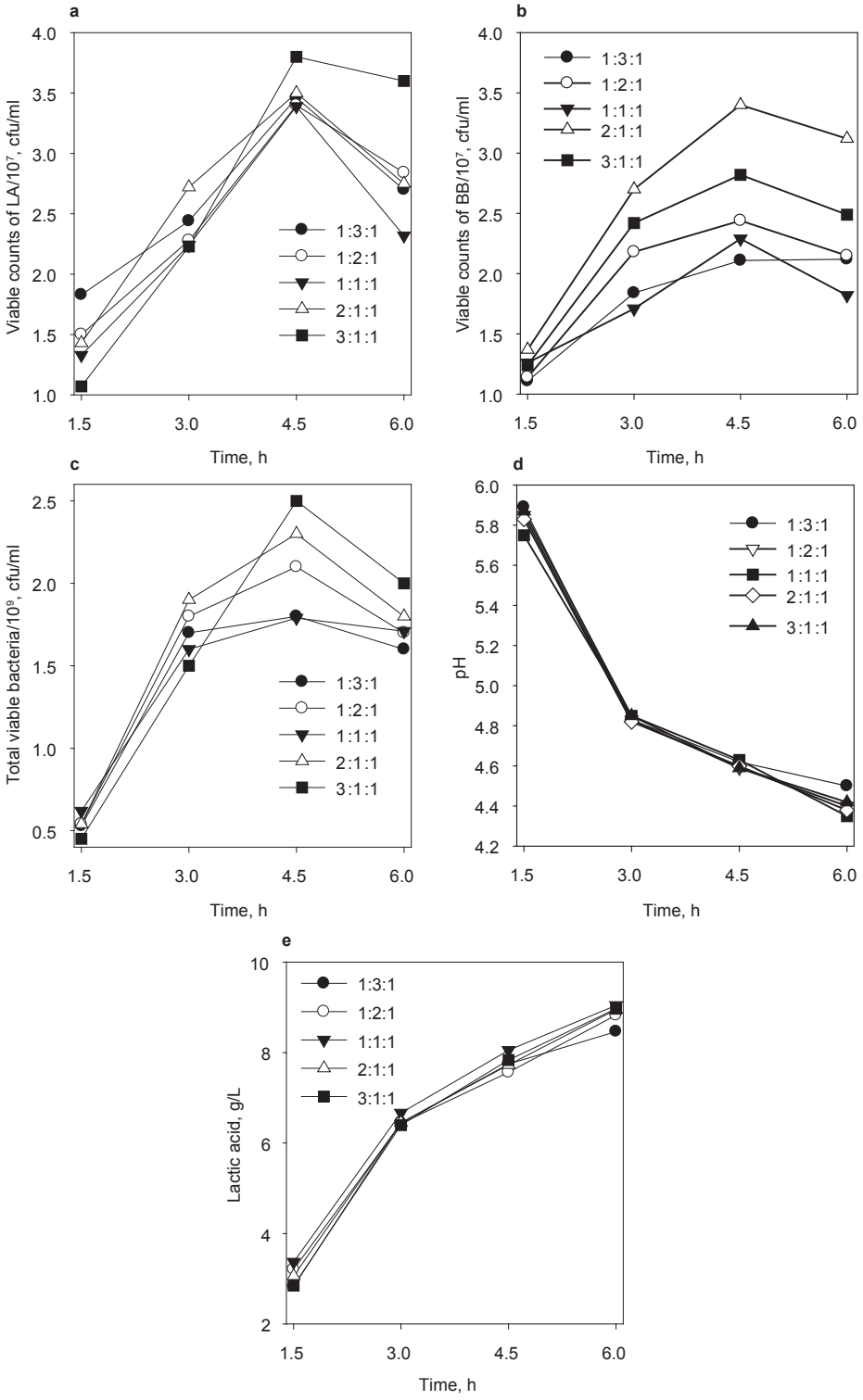


**Fig. 1.** Effect of bacteria proportion on viable counts of *L. acidophilus* and *B. bifidum*, total viable bacteria, pH and acidity in AB-goat yoghurt

Table 1 showed the bacteria proportion had no obvious influence on the colour, texture of goat yoghurt, however, it had a significant effect on goat yoghurt odour and taste. Among them, the sour of goat yoghurt taste a little pale and has slight goaty flavor at 1:1:1, 1:2:1, 1:3:1, 3:1:1 bacteria proportion. The sweet and sour of goat yoghurt was moderate, goaty flavour and comprehensive evaluation was better in 2:1:1 bacteria proportion.

The changes of the total viable counts, *L. acidophilus* and *B. bifidum* in AB-goat yoghurt were all first increased then decreased, the pH decreased and acidity increased during the fermentation process, which affected the growth and survival of probiotics in goat yoghurt. According to sensory evaluation, viable counts, acidity and pH of goat milk fermented by probiotics, the optimum bacteria proportion *L. acidophilus* : *B. bifidum* : *S. thermophilus* and *L. bulgaricus* for AB-goat yoghurt was 2:1:1.

## Effect of bacteria proportion on the fermentation of BC-goat yoghurt

The effect of bacteria proportion on the fermentation of BC-goat yoghurt was shown in Figure 2 and Table 2.

Figure 2a showed *B. bifidum* grew faster in BC-goat yoghurt at 2:1:1 and 3:1:1 than other bacteria proportion. The viable counts of *B. bifidum* increased at 1:3:1 bacteria proportion, first increased within 4.5 h, and then decreased between 4.5 h and 6.0 h at 1:2:1, 2:1:1 and 3:1:1 bacteria proportion, first increased within 3.0 h, and then decreased between 3.0 h and 6.0 h at 1:1:1 bacteria proportion with time increasing. The viable counts of *B. bifidum* in BC-goat yoghurt samples increased from $1.10\times10^7$ cfu/ml at 1.5 h to $3.20\times10^7$ cfu/ml at 6.0 h at 1:3:1 bacteria proportion, first increased from $1.40\times10^7$ cfu/ml at 1.5 h to $3.41\times10^7$ cfu/ml at 4.5 h, and then decreased to $2.52\times10^7$ cfu/ml at 6 h when the bacteria proportion was 1:2:1, first increased from $2.60\times10^7$ cfu/ml at 1.5 h to $3.10\times10^7$ cfu/ml at 3.5 h, and then decreased to $2.21\times10^7$ cfu/ml at 6 h when the bacteria proportion was 1:1:1, first increased from $3.31\times10^7$ cfu/ml at 1.5 h to $7.60\times10^7$ cfu/ml at 4.5 h, and then decreased to $6.70\times10^7$ cfu/ml at 6 h when the bacteria proportion was 2:1:1, first increased from $4.00\times10^7$ cfu/ml at 1.5 h to $7.90\times10^7$ cfu/ml at 4.5 h, and then decreased to $6.30\times10^7$ cfu/ml at 6 h when the bacteria proportion was 3:1:1. Among then, the viable counts of *B. bifidum* at 3:1:1 bacteria proportion reached the maximum ($7.90\times10^7$ cfu/mL) and followed at 2:1:1 ($7.60\times10^7$ cfu/mL), the viable counts of *B. bifidum* at 1:1:1 bacteria proportion was the lowest ($2.90\times10^7$ cfu/ml) at 4.5 h.

Figure 2b showed the viable counts of *L. casei* in all BC-goat yoghurt samples first increased within 4.5 h, and then decreased between 4.5 h and 6.0 h. The viable counts of *L. casei* increased slowly to reach maximum at 4.5 h. Among them, the viable counts of *L. casei* at 2:1:1 bacteria proportion reached the maximum ($5.60\times10^7$ cfu/mL), the viable counts of *L. casei* at 1:1:1 bacteria proportion was the lowest ($3.90\times10^7$ cfu/ml) at 4.5 h.

Figure 2c showed the total viable counts in BC--goat yoghurt were all first increased then decreased

**Table 2.** Sensory evaluation of BC-goat yoghurt fermented by different bacteria proportion

| Bacteria proportion of BB:LC:LB + ST | Colour | Smell | Taste | Texture | Comprehensive evaluation |
|---|---|---|---|---|---|
| 1:3:1 | 1.00 | 2.26 | 1.87 | 2.40 | 7.53 |
| 1:2:1 | 1.00 | 2.29 | 1.97 | 2.41 | 7.67 |
| 1:1:1 | 1.00 | 2.36 | 1.97 | 2.40 | 7.73 |
| 2:1:1 | 1.00 | 2.40 | 1.98 | 2.43 | 7.81 |
| 3:1:1 | 1.00 | 2.44 | 1.92 | 2.37 | 7.73 |

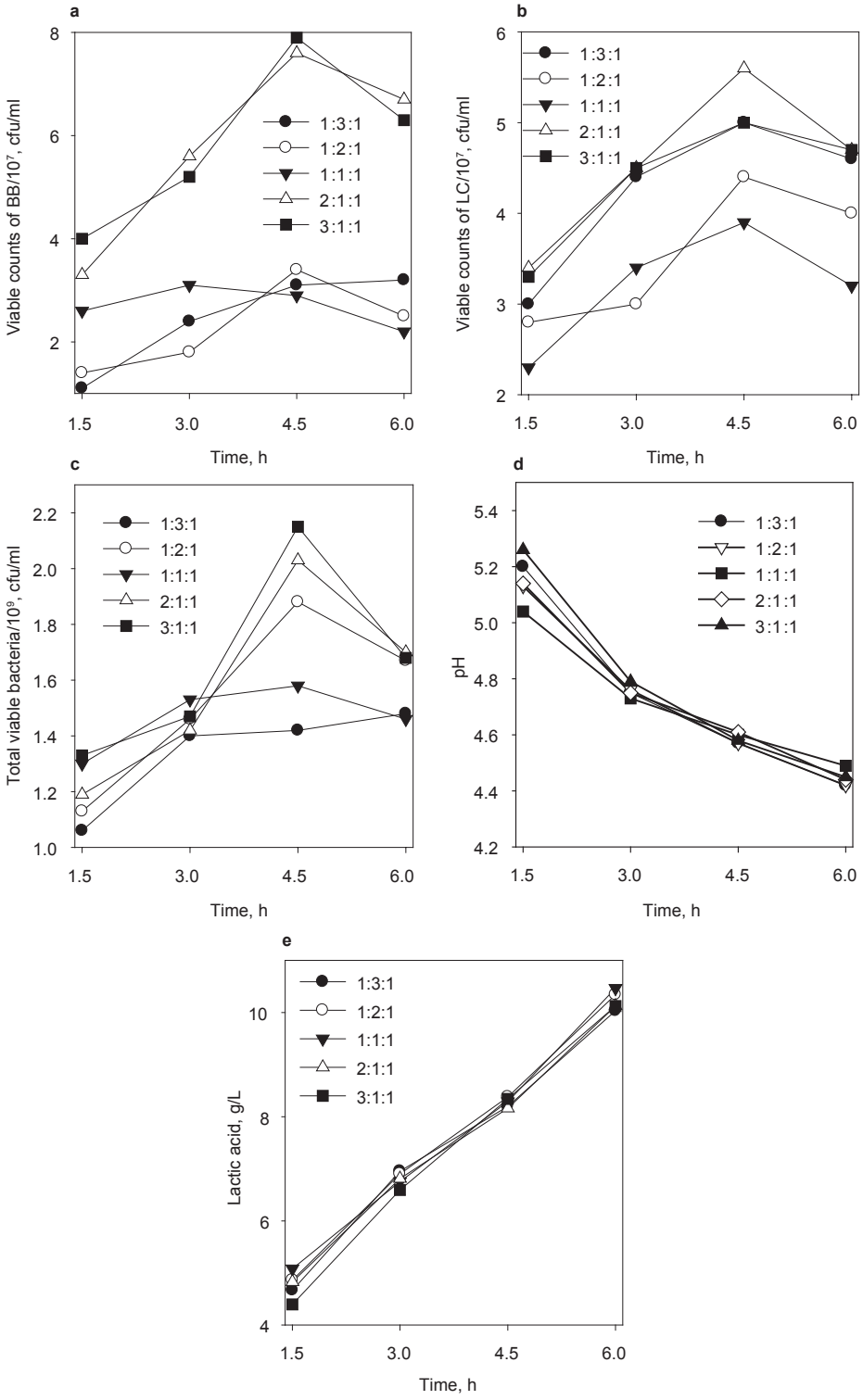


**Fig. 2.** Effect of bacteria proportion on viable counts of *B. bifidum* and *L. casei*, total viable bacteria, pH and acidity in BC-goat yoghurt

except at 1:3:1 bacteria proportion with time increasing. Among them, the total viable counts at 3:1:1 bacteria proportion reached the maximum ($2.15\times10^9$ cfu/mL) followed at 2:1:1 ($2.04\times10^9$ cfu/mL), the total viable counts at 1:3:1 bacteria proportion was the minimum ($1.42\times10^9$ cfu/mL) at 4.5 h. From Figure 2d and 1e, the pH decreased and the acidity increased in all AB-goat yoghurt. The bacteria proportion had no significant influence on acidity and pH ($p < 0.05$).

Table 2 showed the bacteria proportion had no significant influence on the colour, taste and texture of BC-goat yoghurt ($p > 0.05$), however, had a significant effect on odour. The BC-goat yoghurt had obvious goaty flavour at 1:2:1 and 1:3:1, but had slight goaty flavour at 1:1:1, 2:1:1, 3:1:1; the comprehensive evaluation at 2:1:1 bacteria proportion was better than the other.

According to the analysis and discussion of the above, the optimum bacteria proportion of *B. bifidum* : *L. casei* : *S. thermophilus* and *L. bulgaricus* for BC--goat yoghurt was 2:1:1.

## CONCLUSIONS

The bacteria proportion had a significant effect on fermentation of AB- and BC-goat yoghurt. The optimum bacteria proportion for AB-goat yoghurt and BC-goat yoghurt were both 2:1:1. The pH, acidity, the viable counts of *L. acidophilus* and *B. bifidum*, the total viable counts were respectively 4.60, 7.73 (g/L), $3.50\times10^7$ cfu/mL, $3.40\times10^7$ cfu/mL and $2.30\times10^9$ cfu/mL in AB-goat yoghurt. The pH, acidity, the viable counts of *B. bifidum* and *L. casei*, the total viable counts were respectively 4.61, 8.16 (g/L), $7.60\times10^7$ cfu/mL, $5.60\times10^7$ cfu/mL and $2.04\times10^9$ cfu/mL in BC-goat yoghurt.

## ACKNOWLEDGEMENT

The project was partly supported by the Science and Technology Research Development plan project of Shaanxi Province, China (No. 2014K01-17-07), the Scientific Research Program Funded by Shaanxi Provincial Education Department (No. 2013JK0747) and the science and technology plan project of Xi'an city [No. NC1317 (1)].

## REFERENCES

Boycheva, S., Dimitrov, T., Naydenova, N., Mihaylova, G. (2011). Quality characteristics of yoghurt from goat's milk, supplemented with fruit juice. Czech J. Food Sci., 29,1, 24–30.

Chen, H., Wang, C., Shu, G. (2010). Technological optimization of set-style goat yoghurt fermentation. Sci. Techn. Food Ind., 35, 12, 71–74.

Chen, H., Ji, L., Shu, G., Wang, Z. (2011). Effect of lithium chloride and sodium propionate on growth of selected probiotics. Key Eng. Mater., 480/481, 66–69.

Chen, H., Zhang, Q., Wan, H., Shu, G., Li, H. (2015). Effect of total inoculum size containing *Lactobacillus acidophilus* or *Lactobacillus casei* on fermentation of goat milk. Adv. J. Food Sci. Techn., 7, 3, 183–186.

Farnsworth, J. P., Li, J., Hendricks, G. M., Guo, M. R. (2006). Effects of transglutaminase treatment on functional properties and probiotic culture survivability of goat milk yoghurt. Small Ruminant Res., 65, 1, 113–121.

Haenlein, G. F. W. (2004). Goat milk in human nutrition. Small Ruminant Res., 51, 2, 155–163.

Kailasapathy, K., Chin, J. (2000). Survival and therapeutic potential of probiotic organisms with reference to *Lactobacillus acidophilus* and *Bifidobacterium* spp. Immunol. Cell Biol., 78, 1, 80–88.

Krasaekoopt, W., Bhandari, B., Deeth, H. (2003). Evaluation of encapsulation techniques of probiotics for yoghurt. Int. Dairy J., 13, 1, 3–13.

Mutlu, B. G., Akın, S. M. (2007). Effects of cysteine and different incubation temperatures on the microflora, chemical composition and sensory characteristics of bio-yoghurt made from goat's milk. Food Chem., 100, 2, 788–793.

Park, Y. W. (1994). Hypo-allergenic and therapeutic significance of goat milk. Small Ruminant Res., 14, 2, 151–159.

Posecion, N. C., Crowe, N. L., Robinson, A. R., Asiedu, S. A. (2005). The development of a goat's milk yoghurt. J. Sci. Food Agric., 85, 11, 1909–1913.

Rolfe, R. D. (2000). The role of probiotic cultures in the control of gastrointestinal health. J. Nutr., 130, 2, 396–402.

Shu, G., Ma, Z., Wang, Z., Chen, H. (2011). Effect of bile and nalidixic acid on growth of selected probiotics. Adv. Mat. Res., 322, 248–251.

Shu, G., Li, C., Chen, H., Wang, C. (2014). Effect of inoculum and temperature on the fermentation of goat yoghurt. Adv. J. Food Sci. Techn., 6, 1, 68–71.

Singh, K., Kallali, B., Kumar, A., Thaker, V. (2011). Probiotics: A review. Asian Pac. J. Tropic. Biomed. 2, 1, S287–S290.

Uysal-Pala, C., Karagul-Yuceer, Y., Pala, A., Savas, T. (2006). Sensory properties of drinkable yoghurt made from milk of different goat breeds. J. Sens. Stud., 21, 5, 520–533.

Received – Przyjęto: 9.04.2015

Accepted for print – Zaakceptowano do druku: 25.07.2015

For citation – Do cytowania

Shu, G., Wang, S., Chen, Z., Chen, H., Wang, C., Ma, Y. (2015). Effect of bacteria proportion on the fermentation of goat yoghurt with probiotic culture. Acta Sci. Pol. Technol. Aliment., 14(4), 407–414. DOI: 10.17306/J.AFS.2015.4.40