

## Microbiological, pH and Sensory Evaluations of Refrigerated Pork Tender-Loins Treated with Potassium Sorbate

Chang-Ryoul Kim<sup>†</sup>

Department of Food and Nutrition Science, Seo Kang College, Kwangju 500-742, Korea

### Potassium Sorbate에 침지한 냉장 돼지고기 안심의 미생물, pH 및 관능평가

김창렬<sup>†</sup>

서강정보대학 식품영양학과

(Received February 7, 2007/Accepted March 21, 2007)

**ABSTRACT** – Microbiological, pH, and sensory evaluations on pork tender-loins treated with potassium sorbate (PS) during storage at 4°C were assessed. Treatments of 0.5-2.0% PS for 10 min effectively ( $P < 0.05$ ) inhibited the growth of aerobic plate counts (APC) and gram-negative bacterial counts (GNC) compared to those of controls for 9 days at 4°C, respectively. Results of this investigation demonstrate that antimicrobial activity on APC and GNC in all treatments enhanced by increasing concentrations of PS during storage at 4°C. pH values of pork tender-loins treated with PS gradually increased by storage of 6 days at 4°C. Sensory data showed that 0.5-1.0% PS treatments were quite close in odor and appearance compared to fresh controls. Results indicated that pork tender-loins treated with 1.0% PS for 10 min could extend shelf-life to 9 days during refrigerated storage at 4°C without adversely affecting sensory quality.

**Key words:** pork tender-loin, potassium sorbate, pH, APC, sensory evaluation

Extending shelf-life of meat depends on maintaining low microbial numbers during storage at refrigeration temperatures, which is important to the meat industry because it is the main factor associated with reduced quality of meat, spoilage, and subsequent economic loss.<sup>1-13</sup> The potassium sorbate (PS) plays a major role in meat and fish processing due to its antimicrobial activity and is often used to extend the shelf-life of many foods.<sup>1,2,6,7,10,11</sup> Studies have shown that PS is used to reduce the level of aerobic food spoilage bacteria and food pathogens in several foods.<sup>1,6,7,11</sup> Mendonca *et al.*<sup>7</sup> reported that vacuum-packaged pork chops treated with PS reduced counts of mesophiles, psychrotrophs, *Enterobacteriaceae*, facultative anaerobes, and lactobacilli. They have shown that combined use of 10% phosphate and 10% PS solutions extended shelf life in vacuum-packaged fresh pork chops to 10 weeks at 2-4°C com-

pared with untreated pork and protected meat colors. EL-Shenawy and Marth<sup>2</sup>) noted that a combination of two preservatives is more effective against undesirable microorganisms in tryptose broth than just one alone. They found that a combination of PS with lactic or acetic acid more effectively inactivated *Listeria monocytogenes* than potassium sorbate by itself. The antimicrobial activity of PS combined with sodium acetate and lactic acid bacteria was demonstrated by Kim and Hearnberger<sup>4</sup>) who observed that growth of gram-negative bacteria was inactivated in refrigerated catfish fillets. Staham *et al.*<sup>6</sup>) have shown that antimicrobial and sensory evaluations on vacuum packaged fillets treated with PS was more effective than a 100% CO<sub>2</sub> atmosphere alone. Kim *et al.*<sup>8</sup>) noted that the decontamination of aerobic spoilage bacteria is essential in meat and its products. Among those microorganisms gram-negative bacteria were the major cause of microbial spoilage in refrigerated meat and fish.<sup>3-5,8</sup>

However, there are limited studies to determine the efficacy of PS for aerobic spoilage bacteria, gram neg-

<sup>†</sup>Correspondence to: Chang-Ryoul Kim, Dept. of Food Science and Nutrition, SeoKang College, Kwangju 500-742, Korea. Tel: +82-62-520-5206, C.P.: 017-605-5186, Fax: +82-62-520-5206, E-mail : changkim@skc.ac.kr

ative bacteria, pH, and sensory evaluations on refrigerated pork tender-loins. The purpose of our study was to evaluate shelf-life extension of refrigerated (4°C) pork tender-loins by treating their surfaces with PS.

## Materials and Methods

### Pork preparation and treatments

Fresh pork tender-loins were obtained from a commercial source and transported to the laboratory on ice and used within 2 h of arrival. Each one  $\beta^3$  of pork tender-loins (average weight 50 g per tender-loins) were placed in a tumbler (Chang Dae Chemical Co., Model 101, Korea) at room temperature. Potassium sorbate (PS) was added to the pork tender-loins and tumbled for 10 min at 15 rpm. PS, at concentrations of 0.5, 1.0, 1.5, and 2.0% (w/w) was used in this work. Controls were tumbled without PS for 10 min at 15. Treated pork tender-loins were packed in groups of four in large Whirl-Pak bags (Fisher Chemical Co., USA) and labelled. The bags were closed and stored at 4°C. Samples were removed from storage at appropriate times for analyses.

### Microbiological analysis

Individual pork tender-loin was aseptically transferred to Whirl-Pak bags, weighed, and diluted 1 : 1 with 0.1% (w/v) sterile peptone water. Samples were shaken 60 times using the standard rinse method (4). The liquid from each sample was diluted and plated in volumes of 0.1 ml on standard plate count agar (Difco Laboratories, Detroit, USA) for aerobic plate counts (APC) and Mac-Conkey agar (Difco Laboratories, Detroit, USA) for gram-negative bacterial counts (GNC), respectively. The plates were incubate for 48 hr at 30°C before counting the colonies. The number of bacteria was expressed as mean  $\text{Log}_{10}$  CFU/g of the duplicate treatments.

### Calculation of growth curves

The growth rates of aerobic microorganisms were used to calculate generation times.<sup>14)</sup> Two points on the logarithmic growth phase of each curve were used in the calculation.

$$\text{Generation time (GT)} = (0.301(T_2 - T_1)) / (\log P_2 - P_1)$$

Where,  $T_1$  = time of  $P_1$ ,  $T_2$  = time of  $P_2$ ,  $P_1$  = CFU/g at  $T_1$ , and  $P_2$  = CFU/g at  $T_2$ .

### pH measurements

pH values of the homogenates of the muscle (2.5 g) in 10 ml of 5 mM iodoacetate-150 mM KCl (adjusted to pH 7.0) were measured according to the method of Bendall.<sup>15)</sup>

### Sensory evaluations

Sensory evaluations of samples were performed by an ten member trained panel. Odor and appearance of uncooked pork tender-loins were evaluated during storage at 4°C. Treated pork tender-loins were judged against fresh control pork tender-loins (fresh daily), which were assigned a score of 5. Samples liked less than the control were scored 1 to 4, where 1 = disliked most. Samples liked more than the control were scored 6 to 9, where 9 = liked most. Untreated control pork tender-loins also were stored at 4°C for comparison against fresh control pork tender-loins and treated pork tender-loins.

### Statistical analyses

APC, GNC, pH, and sensory data were analyzed using ANOVA, and means were separated by the least significant difference test at  $P < 0.05$  using SAS program.<sup>16)</sup>

## Results and Discussion

### Microbiological analysis

APC and GNC on pork tender-loins treated with different levels of PS for 10 min using a tumbler at room temperature were significantly ( $P < 0.05$ ) lower than the controls during storage at 4°C (Table 1 and 2). Treatments of 1.0-2.0% (w/w) PS were effective in lowering the initial level of APC and GNC. Increasing concentrations of PS by 1.0 to 2.0% (w/w) significantly ( $P < 0.05$ ) inhibited the growth of aerobic spoilage bacteria and gram negative bacteria in pork tender-loins for 9 days of storage compared to 0.5% PS treatments, while the control pork tender-loins were rapidly spoiled as evidenced by the microbial growth. Mendonca *et al.*<sup>7)</sup> have shown that potassium sorbate alone was significantly ( $P < 0.05$ ) more effective in delaying the growth of lactobacilli in vacuum-packaged pork chops than it was in combination with other chemicals. They suggested that treatment of 10% Brifisol 414<sup>TM</sup> without PS did not contribute extended shelf life in vacuum-packaged pork

**Table 1. APC values\* on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS)**

Treatment	Storage time (days)			
	Log CFU/g			
	0	3	6	9
Control	3.46 ± 0.18 <sup>a</sup>	3.58 ± 0.11 <sup>a</sup>	5.47 ± 0.12 <sup>a</sup>	7.41 ± 0.17 <sup>a</sup>
0.5%PS/15 rpm	3.50 ± 0.06 <sup>a</sup>	3.71 ± 0.08 <sup>a</sup>	4.41 ± 0.23 <sup>b</sup>	5.56 ± 0.10 <sup>b</sup>
1.0%PS/15 rpm	2.91 ± 0.05 <sup>ab</sup>	2.80 ± 0.04 <sup>b</sup>	2.72 ± 0.09 <sup>c</sup>	2.96 ± 0.06 <sup>c</sup>
1.5%PS/15 rpm	2.45 ± 0.17 <sup>b</sup>	2.72 ± 0.21 <sup>b</sup>	2.70 ± 0.08 <sup>c</sup>	2.70 ± 0.05 <sup>c</sup>
2.0%PS/15 rpm	2.38 ± 0.18 <sup>b</sup>	2.50 ± 0.11 <sup>b</sup>	2.26 ± 0.04 <sup>c</sup>	2.32 ± 0.13 <sup>c</sup>

\*Mean ± SD within the same column with different superscripts are significantly different (P < 0.05).

**Table 2. GNC values\* on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS)**

Treatment	Storage time (days)			
	Log CFU/g			
	0	3	6	9
Control	3.21 ± 0.04 <sup>a</sup>	2.99 ± 0.09 <sup>a</sup>	4.57 ± 0.28 <sup>a</sup>	5.62 ± 0.02 <sup>a</sup>
0.5%PS/15 rpm	3.34 ± 0.02 <sup>a</sup>	3.35 ± 0.05 <sup>a</sup>	3.58 ± 0.43 <sup>bc</sup>	2.45 ± 0.11 <sup>b</sup>
1.0%PS/15 rpm	2.25 ± 0.07 <sup>b</sup>	2.12 ± 0.05 <sup>b</sup>	2.06 ± 0.13 <sup>cd</sup>	1.94 ± 0.24 <sup>b</sup>
1.5%PS/15 rpm	2.05 ± 0.18 <sup>b</sup>	1.79 ± 0.29 <sup>b</sup>	1.67 ± 0.13 <sup>d</sup>	1.50 ± 0.00 <sup>bc</sup>
2.0%PS/15 rpm	2.13 ± 0.07 <sup>b</sup>	1.72 ± 0.14 <sup>b</sup>	1.57 ± 0.13 <sup>d</sup>	1.00 ± 0.07 <sup>c</sup>

\*Mean ± SD within the same column with different superscripts are significantly different (P < 0.05).

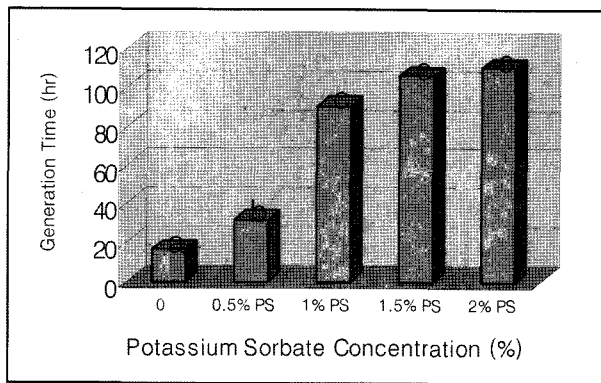
chops. Moreover, PS was considered important because the lactobacilli have been recognized as the predominant spoilage microbial group in vacuum-packaged, refrigerated, fresh or processed meats. Statham *et al.*<sup>6)</sup> have reported that Morwong (*Nemadactylus macropterus* Bloch and Schneider) fillets treated with combination of potassium sorbate, polyphosphate and 100% CO<sub>2</sub> atmosphere packaging were the most effective packaging regime. PS on vacuum packaged fish was more effective than a 100% CO<sub>2</sub>. They reported that polyphosphate had no apparent additional effect on fillets stored under vacuum with or without potassium sorbate. Furthermore, growth of the food spoilage bacteria and food pathogens such as gram negative bacteria, aerobic mesophiles, psychrotrophic bacteria, anaerobic and facultative anaerobic bacteria, and *Listeria monocytogenes* in meat and fish was inhibited to various degrees by combinations of PS and other chemicals.<sup>2,6,7,11)</sup> EL-Shenawy and Marth<sup>2)</sup> noted that a combination of PS and acid had a greater antimicrobial activity in tryptose broth than that of either alone. They found that the antilisterial effect of potassium sorbate was enhanced by the presence of organic acids such as lactic acid, acetic acid, or citric acid. Kim and Hearnberger<sup>4)</sup> noted that treatments of 0.5% (w/w) PS alone or 0.25% PS combined with both 0.50%

sodium acetate and 2.5% lactic acid cultures (*Lactococcus lactis* ATCC 19257) had a significantly lower levels of GNC on fresh fish fillets during storage of 10 days at 4°C. The combination of 0.25% PS and 0.50% sodium acetate with 2.5% lactic acid cultures or 1.0% sodium acetate, alone, improved catfish odor and appearance during 13 days of storage.

Results indicated that microbial spoilage of pork tender-loins is mainly caused by gram negative bacteria, which were sensitive to increasing levels of PS. Darmadji and Izumimoto<sup>17)</sup> noted that gram-negative bacteria such as *Pseudomonas* spp. secrete proteolytic enzymes which play a role in the degradation of protein. They noted that this process may affect meat putrefaction and production of volatile nitrogen. This suggested that a use of PS as a sanitizer could be necessary to extend a microbiological shelf-life on pork meat as Mendonca *et al.*<sup>7)</sup> found it to be in vacuum-packaged pork chops. Although PS has an antimicrobial effect in pork tender-loins, it is recommended to use in food by 25mg/kg/wt.

#### Analysis of generation time

When pork tender-loins treated with 0.5-2.0% (w/w) PS for 10 min, 1.0% PS increased generation times by



**Fig. 1.** Generation time of aerobic plate counts (APC) on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS). Means sharing the same letter are not significantly different ( $P > 0.05$ ).

90.3 hr (Fig. 1). There was significantly different ( $P < 0.05$ ) on GT between controls and treatments. Treatment of 1.0% PS was no significantly different ( $P > 0.05$ ) on GT compared to that of other treatments. PS not only caused a decrease in the initial number of aerobic spoilage bacteria (Table 1 and 2) but also decreased the growth rate of survivors as reflected by increasing generation times (Fig. 1). Results indicate that the effectiveness of PS as an antimicrobial surface treatment attributed to increasing levels of PS by 2.0%.

### pH values

pH of the pork tender-loins significantly ( $P < 0.05$ ) increased initially due to PS (Table 3). All pH values of pork tender-loins treated with PS gradually increased by

**Table 3.** pH values\* on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS)

Treatment	Storage time (days)			
	0	3	6	9
Control	5.56 ± 0.01 <sup>b</sup>	5.64 ± 0.01 <sup>b</sup>	5.89 ± 0.02 <sup>b</sup>	6.21 ± 0.11 <sup>a</sup>
0.5% PS <sup>2</sup> /15 rpm	6.04 ± 0.02 <sup>a</sup>	6.02 ± 0.02 <sup>a</sup>	6.10 ± 0.01 <sup>a</sup>	6.05 ± 0.02 <sup>a</sup>
1.0% PS/15 rpm	6.06 ± 0.01 <sup>a</sup>	6.07 ± 0.02 <sup>a</sup>	6.08 ± 0.02 <sup>a</sup>	6.09 ± 0.01 <sup>a</sup>
1.5% PS/15 rpm	6.09 ± 0.02 <sup>a</sup>	6.05 ± 0.03 <sup>a</sup>	6.09 ± 0.02 <sup>a</sup>	6.11 ± 0.02 <sup>a</sup>
2.0% PS/15 rpm	6.09 ± 0.03 <sup>a</sup>	6.08 ± 0.01 <sup>a</sup>	6.06 ± 0.02 <sup>a</sup>	6.06 ± 0.02 <sup>a</sup>

\*Mean ± SD within the same column with different superscripts are significantly different ( $P < 0.05$ ).

**Table 4.** Odor values\* on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS)

Treatment	Storage time (days)			
	0	3	6	9
Control	5.00 ± 0.32 <sup>b</sup>	4.60 ± 0.25 <sup>b</sup>	3.80 ± 0.20 <sup>b</sup>	3.60 ± 0.25 <sup>b</sup>
0.5% PS <sup>2</sup> /15 rpm	5.60 ± 0.40 <sup>ab</sup>	5.20 ± 0.37 <sup>a</sup>	5.00 ± 0.55 <sup>a</sup>	4.60 ± 0.25 <sup>a</sup>
1.0% PS/15 rpm	5.40 ± 0.25 <sup>ab</sup>	5.20 ± 0.20 <sup>a</sup>	4.60 ± 0.25 <sup>a</sup>	4.60 ± 0.40 <sup>a</sup>
1.5% PS/15 rpm	6.00 ± 0.45 <sup>a</sup>	5.40 ± 0.25 <sup>a</sup>	5.00 ± 0.32 <sup>a</sup>	3.20 ± 0.20 <sup>b</sup>
2.0% PS/15 rpm	5.60 ± 0.68 <sup>a</sup>	4.80 ± 0.49 <sup>b</sup>	4.80 ± 0.49 <sup>a</sup>	3.60 ± 0.25 <sup>b</sup>

\*Mean ± SD within the same column with different superscripts are significantly different ( $P < 0.05$ ).

**Table 5.** Appearance values\* on refrigerated (4°C) pork tender-loins treated with different levels of potassium sorbate (PS)

Treatment	Storage time (days)			
	0	3	6	9
Control	5.40 ± 0.40 <sup>b</sup>	4.60 ± 0.25 <sup>b</sup>	4.40 ± 0.25 <sup>b</sup>	3.80 ± 0.20 <sup>b</sup>
0.5% PS <sup>2</sup> /15 rpm	5.40 ± 0.60 <sup>b</sup>	5.40 ± 0.40 <sup>ab</sup>	4.40 ± 0.40 <sup>b</sup>	4.20 ± 0.37 <sup>a</sup>
1.0% PS/15 rpm	6.60 ± 0.40 <sup>ab</sup>	5.60 ± 0.40 <sup>ab</sup>	4.80 ± 0.49 <sup>ab</sup>	4.80 ± 0.20 <sup>a</sup>
1.5% PS/15 rpm	7.40 ± 0.51 <sup>a</sup>	6.80 ± 0.20 <sup>a</sup>	5.20 ± 0.37 <sup>a</sup>	4.20 ± 0.58 <sup>a</sup>
2.0% PS/15 rpm	7.60 ± 0.25 <sup>a</sup>	6.20 ± 0.20 <sup>a</sup>	5.40 ± 0.25 <sup>a</sup>	4.60 ± 0.25 <sup>a</sup>

\*Mean ± SD within the same column with different superscripts are significantly different ( $P < 0.05$ ).

storage of 6 days at 4°C. However, pH values of pork tender-loins treated with PS during storage of 9 days at 4°C were no significant difference ( $P < 0.05$ ) compared to those of controls. Results showed that pH values relating PS had no direct antimicrobial effect on the reduction of aerobic food spoilage bacteria on pork tender-loins. Mendonca *et al.*<sup>7)</sup> have shown that treatments of phosphate blend-PS combinations or its alone increased pH values in refrigerated pork chops. They suggested that the microbiological effects of treatments were a relationship with solution uptake, exudate, and pH of phosphate blend by pork chops. Moreover, Kim and Hearnshberger<sup>4)</sup> noted that the pH values on catfish fillets treated with 0.5% (w/w) PS alone or 0.25% PS combined with both 0.50% sodium acetate and 2.5% lactic acid cultures (*Lactococcus lactis* ATCC 19257) drop to 6.40 in 9 days at 4°C. They suggested that the antimicrobial effects of PS and other chemicals did not appear to be due to pH values.

### Sensory analysis

Sensory data of pork tender-loins treated in 0.5-2.0%

(w/w) PS for 10 min are shown in Table 4 and 5. Sensory scores indicated that PS treatments were in the "liked less to typical" category in odor and appearance compared to the fresh controls. Odor and appearance scores were statistically distinguishable ( $P < 0.05$ ) from 1.5% PS treatments at initial day. Appearance scores were significantly lower ( $P < 0.05$ ) from 0.5-2.0% PS treatments during storage of 9 days. Refrigerated pork tender-loins treated with 0.5-1.0% PS were quite close in odor and appearance scores to fresh controls through the storage time. Similarly, Kemp *et al.*<sup>1)</sup> have shown that PS had no effect on yield but dry PS generally improved panel scores in boneless pork hams. The sensory data indicate that PS concentrations up to 1.0% could be used on refrigerated pork tender-loins without contributing a typical chemical odor and discoloration.

### Acknowledgements

This study was supported by a grant of the ARPC project, Ministry of Agriculture and Forestry.

### 국문요약

본 연구는 4°C 냉장 동안 potassium sorbate로 침지한 돼지고기 안심의 미생물, pH 및 관능평가를 실시하였다. 0.5~2.0% (w/w) potassium sorbate로 10분 동안 침지한 돼지고기 안심의 호기성 미생물과 그람음성 미생물은 대조군과 비교하여 4°C 냉장 9일 동안 유의적 감소( $P < 0.05$ )를 보였다. 본 연구의 결과 4°C 냉장 동안 potassium sorbate의 농도가 증가하므로써 모든 처리군에서 돼지고기 안심의 호기성 미생물과 그람음성 미생물에 대한 항미생물 효과를 보였다. 4°C 냉장 6일 동안 potassium sorbate로 침지한 돼지고기 안심의 pH기는 증가함을 보였다. 0.5~1.0% (v/w) potassium sorbate로 침지한 돼지고기 안심의 관능평가 결과는 신선한 대조군과 비교하여 냄새와 외관의 점수가 매우 유사함을 보였다. 본 연구의 결과 1.0% (v/w) potassium sorbate로 10분 침지한 돼지고기 안심은 관능평가 저하에 영향을 미치지 않고 4°C 냉장 동안 미생물학적 저장 안정성을 확장할 수 있다는 것을 입증하였다.

### References

1. Kemp, J.D., Langlois, B.E., and Fox, J.D. Effect of potassium sorbate and vacuum packaging on the quality and microflora of dry-cured intact and boneless hams. *J. Food Sci.*, **48**: 1709-1712 (1983).
2. EL-Shenawy, M.A. and Marth, E.H. Organic acids enhance the antilisterial activity of potassium sorbate. *J. Food Prot.*, **54**: 593-597 (1991).
3. Kim, C.R. and Marshall, D.L. Microbiological, colour and sensory changes of refrigerated chicken legs treated with selected phosphates. *Food Research International*, **32**: 209-215 (1999).
4. Kim, C.R. and Hearnshberger, J.O. Gram negative bacteria inhibition by lactic acid culture and food preservatives on catfish fillets during refrigerated storage. *J. Food Sci.* **59**: 513-516 (1994).
5. Kim, C.R. and Kim, K.H. Physicochemical quality and

- gram negative bacteria in refrigerated chicken legs treated with trisodium phosphate and acetic acid. *Food Sci. Biotech.* **9**: 218-221 (2000).
6. Statham, J.A., Bremner, H.A., and Quarmby, A.R. Storage of Morwong (*Nemadactylus macropterus* Bloch and Schneider) in combination of polyphosphate, potassium sorbate and carbon dioxide at 4°C. *J. Food Sci.* **50**: 1580-1587 (1985).
  7. Mendonca, A.F., Molins, R.A., Kraft, A.A., and Walker, H.W. Effects of potassium sorbate, sodium acetate, phosphates and sodium chloride alone or in combination on shelf life of vacuum-packaged pork chops. *J. Food Sci.* **54**: 302-306 (1989).
  8. Kim, C.R., Lee, J.I., Kim, K.H., Kang, C.K., Rhie, S.C., Moon, S.J., and Lee, Y.K. Microbiological and sensory evaluations of refrigerated pork loins treated with citric acid. *Korean Vet. Publ. Hlth.* **20**: 329-335 (1996).
  9. Mendonca, A.F., Molins, R.A., Kraft, A.A., and Walker, H.W. Microbiological, chemical and physical changes on fresh, vacuum-packed pork treated with organic acids and salts. *J. Food Sci.* **54**: 18-21 (1989).
  10. Tsay, W.I. and Chou, C.C. Influence of potassium sorbate on the growth of *Yersinia enterocolitica*. *J. Food Prot.* **52**: 723-726 (1989).
  11. Gilland, S.E. and Ewell, H.R. Influence of combinations of *Lactobacillus lactis* and potassium sorbate on growth of psychrotrophs in raw milk. *J. Dairy Sci.* **66**: 974-980 (1983).
  12. Ray, B. and Sandine, W.E. Acetic, propionic, and lactic acids of starter culture bacteria as biopreservatives. In *Food Biopreservatives of Microbial Origin*, Ray, B. and Daeschel, M., CRC Press, Inc., Boca Raton, FL, USA, pp. 103-106 (1991).
  13. Rathgeber, B.M. and Waldroup, A.L. Antibacterial activity of a sodium acid pyrophosphate product in chiller water against selected bacteria on broiler carcasses. *J. Food Prot.* **58**: 530-534 (1995).
  14. Marshall, D.L. and Schmidt, R.H. Growth of *Listeria monocytogenes* at 10°C in milk preincubated with selected pseudomonads. *J. Food Prot.* **51**: 277-282 (1988).
  15. Bendall, J.R. Postmortem changes in muscle. In: G. H. Bourine Ed.): *The structure and function of muscle*, Vol. 2 (2nd Ed.), Academic Press, New York, USA (1994).
  16. SAS. SAS User's Guide. Stastics. SAS Institute Inc., Cary, N. C (1996).
  17. Darmadji, P. and Izumimoto, M. Effect of chitosan in meat preservation. *Meat Sci.* **41**: 243-248 (1994).