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Effect of lactic acid pretreatment on the quality of fresh pork packed in modified atmosphere

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Abstract

Fresh pork ham portion collected from the local supermarket of Wuxi was sprayed with 1, 2, 4 and 6% lactic acid solution at an amount 2 ml per 100 gms, and packed in modified atmosphere of 45% O_2 and 20% CO_2. The samples analyzed during 4th, 8th and 12th days revealed that the color scores and *a** values decreased with increasing concentration of lactic acid and the days of storage. Lower concentration lactic acid treated samples such as 1 and 2%, did not make significant differences with the 0% lactic acid treated control which had color score of 6.9 and *a** value 7.83 on 12th day's analysis. Thiobarbituric acid reactive substances (TBARS) values of the samples increased with the increasing concentration of lactic acid and the days of storage. Lactic acid treated samples at 4 and 6% had higher TBARs values equal to 0.548 and 0.642 mg malonaldehyde/kg meat respectively on 12th day analysis. Four and six percent lactic acid treatments controlled microbial load more significantly among the treatments, which were found to be 5.17 and 5.04 log cfu/gm respectively on 12th day analysis. However, these samples could not keep their color and *a** values stable. In overall, 4 or 6% lactic acid spray treatment could be better if color and lipid oxidation could be stabilized by appropriate stabilizers.

Keywords: Fresh pork; Lactic acid; Modified atmosphere; Lipid oxidation

1. Introduction

Quality of Fresh meat packaged in modified atmosphere depends largely on the initial bacterial load (Christopher, Seideman, Carpenter, Smith, & Vanderzant, 1979). Therefore, various decontamination procedures are being applied in the industries. Organic acid treatment is one of them. Organic acids depending upon their chemical nature vary in their antibacterial properties, while the acceptability of the product also influences on the selection of the acids (Doores, 1993; Brody, 2000). Lactic acid which does not have undesirable odor forming effect even at 5% level application in lamb carcass (Smulders, 1987), is one of the widely used acid decontaminants in meat industry.

The use of lactic acid to decontaminate for the carcass in the earlier period (1–2 days after slaughter) is better than in the later period (more than 2 days after slaughter) because the effect of lactic acid on the color might not be severe at early higher pH and also the bacterial load which needs to be lowered would also be lower (Firstenberg-Eden, 1981). The initial microflora would not get chance to multiply if it is treated earlier, and also the organisms are more resistant to organic acid when the time gap between acid treatment and slaughter is increased (Snijders, Van logtestijn, Mossel, & Smulders, 1985). And also the bacterial number and its maturity are lower in the beginning (Baird-Parker, 1980). On the other hand, the sufficient post mortem

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storage of the meat (24 h or more) and then application of lactic acid could cause severe drop in pH which eventually imparts adverse effect on the color stability of the meat. Sufficient literatures are not available to forecast accurately about the effectiveness of applying lactic acid earlier or at later time, but it is frequently stated that the early application of the lactic acid is more effective than the later treatment (Smulders, 1987; Bell, 2001).

It has been reported that the effect of lactic acid in the reduction of some pathogenic organisms such as Salmonella (Van Netten, Mossel, & Huis In't Veld, 1995) and Campylobacter spp. (Epling, Carpenter, & Blankenship, 1993). It has potential to reduce the microbial load to some extent (Beimuller, Carpenter, & Reynolds, 1973). It has been tested for the fresh beef. However, the use of lactic acid decontamination of the fresh pork chops (individual cuts) has not been carried out yet, especially the meat without allowing 24 h postmortem storage. It is expected that the acid treatment can improve the microbial safety and reduce the spoilage organisms leading to enhanced shelf life in modified atmosphere.

Therefore, the objective of this work is to find the appropriate concentration of lactic acid for decontamination pretreatment of the fresh pork cuts packed in the modified atmosphere and to observe the effect on color and lipid oxidation.

2. Materials and methods

2.1. Raw materials

Fresh pork, a local famous variety of Zhajiaosanhao, ham portion without skin, was collected from the local supermarket of Wuxi, early in the morning, approximately 2 h after slaughter. The meat portion was selected on the basis of color appearance, and the pH was measured by pH paper (range 5.4–7.0) so as to avoid the purchase of PSE meat. The meat which visually seems to have higher myoglobin content (a bit redder in appearance), was purchased in a batch of total 5 kg, 1–2 kg for each chop. It was immediately brought to laboratory and kept in the refrigerator adjusted at 4 °C.

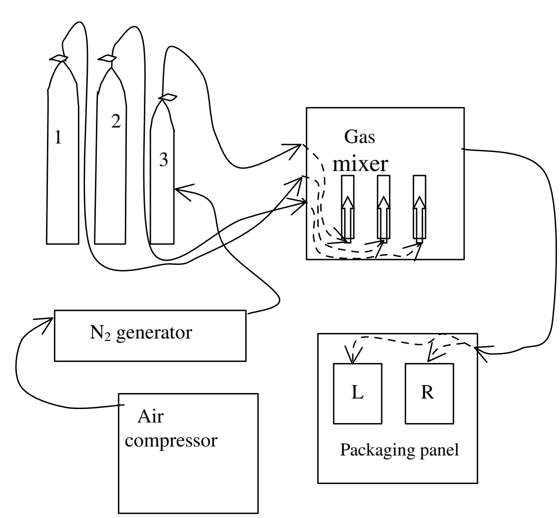
2.2. Packaging and storage procedure

Meat chops in a thickness of 1.5 cm and weight 125 gms for MAP were prepared approximately 6 h after the slaughter. The chops were placed in clean trays. It was then sprayed with 0%, 1%, 2%, 4% and 6% lactic acid solution prepared from sterilized distilled water as diluents. The solution with 85% concentration of lactic acid (Sigma Company) was sprayed with a simple plastic nozzle spray which sprays 1.25 ml at a time. After spray of lactic acid the surface was rubbed in two sides from the centre using a sterilized glass rod so that it would distribute more uniformly. The treated meat containing 2 ml of sprayed solution per 100 gm was packed without delay in the modified atmosphere initially containing 45% O_2 along with 20% CO_2 in each package, where nitrogen was used as a filler gas. The gas to meat ratio adjusted was 3:1 (ml/gm) in the tray of volume 500 ml. The O_2 and CO_2 measuring device (CYES-II Shanghai, ±0.1%) was used for determination of O_2 contents. The packaging was carried out by using Heng Zhong Machine (Fig. 1). Both the plastic tray (CT based) and wrapping plastic film (OPP based), supplied by Zhang Jia Gang Company, used were high barrier to the moisture and gas. Then, the packages were stored in the air-circulated refrigerator at 4 °C for up to 12 days (commercial requirement in China). The process flow chart is mentioned in the Fig. 2.

2.3. Analysis methods

Samples were analyzed for total plate count (TPC), thiobarbituric acid reactive substances (TBARs), color acceptability scores, pH, and *a** value.

(1) *Total plate count (TPC) determination:* On the day of analysis, the samples were opened in the super clean inoculation chamber for microbial tests. The packages were opened aseptically and twenty grams of the representative sample was cut, weighed and transferred to the blender which was previously sterilized with alcohol and rinsed with sterilized distilled water. Onward, 180 ml of phosphate buffer as a diluent (at 4 °C) was added to the blender cup and mixed well at medium speed for 1–2 min to make uniform slurry. The mixture was allowed to settle for a while, less than 10 min. It was serially



Legend: 1= CO_2, 2= O_2, 3= N_2, Gas= gas mixer, L= left, R= right, Packaging = Packaging panel

Fig. 1. Schematic diagram of modified atmospheric packaging machine.

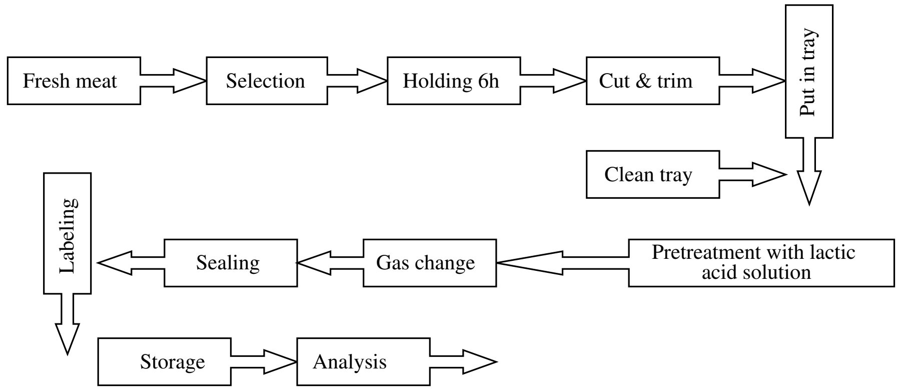


Fig. 2. Flow chart for packaging of fresh pork in MAP with lactic acid pretreatment.

diluted and transferred to sterilized Petri dishes and then plated for total aerobic plate count. It was incubated at 30 °C for 72 h. (Refai, 1979) TPC value is expressed in $\log_{10}$ colony forming units per gram of meat ($\log_{10}$ cfu/gm) throughout this text.

(2) *pH determination:* Ten grams of homogeneous sample was taken in the beaker and then mixed with 100 ml of deionized water and left it for 30 min with periodic stirring and then pH value was measured (Horwitz, 1980) using pH meter (320-S PH equipment, Mettler-Toledo instruments Shanghai Ltd) which had been standardized using standard buffer solutions of pH 4, 9 and 7 chemical mixtures supplied by Shanghai Aijian reagent company Ltd.

(3) *Thiobarbituric acid reactive substances (TBARS) value determination:* The samples were kept in the refrigerator till it was taken for analysis. The samples were first chopped thoroughly using knife and chopping board, and then homogenous sample was made using the warring blender. Twenty grams of the meat sample was mixed with 50 ml of 20% trichloroacetic acid in 2 M phosphoric acid solution at 4 °C, and it was homogenized by vertex breaker homogenizer for 1.5 min. It was diluted with deionized water to make 100 ml and then filtered. 5 ml of the filtrate was mixed with 5 ml of freshly prepared 0.005 M thiobarbituric acid solution in a stopper fitted glass tube. It was mixed simply by inverting the tube several times and then kept in the dark for 15 h at room temperature. Finally, the absorbance of the color developed was measured at 530 nm using UV visual spectrophotometer (752 Leng Guang spectrophotometer; Shanghai Exact Science Equipment Ltd.).

The TBARS value was calculated as following:

$$\text{TBARS value} = [(\text{absorbance} - 0.0121)/0.1379] \times [72.06/94] \text{ mg MDA/kg meat} \quad (1)$$

where, Percent recovery (directly adopted from Witte, Krause, & Bailey, 1970) = 94% and 'MDA' represents malondialdehyde which has a molecular weight of 72.06.

(4) *Instrumental color measurement, a* value:* The measurement of surface color of raw material and thermal treated samples were carried out by using TCP II (Shanghai Optical Instrument Company, China) spectrocolorimeter with white background. The instrument was a tristimulus colorimeter which measures four specific wavelengths in the visible range, specified by the Commission Internationale del' Esclairage (CIE). The surface color of the meat sample at five different places were measured in terms of L, a^* and b^* values using color difference meter. Samples were placed in a special cup, which fits well with the sample port of the colorimeter, to protect it from the interference of outside light.

(5) *Sensory color and overall acceptability scores:* Fifty students who have the knowledge of food science and sensory quality of food are requested to score the color and overall (general appearance color, and wetness or dryness, and odor) acceptability on the basis of nine point hedonic rating scales. The scales include 1 = extremely unacceptable, 2 = Very much unacceptable, 3 = moderately unacceptable, 4 = slightly unacceptable, 5 = between acceptable and unacceptable, 6 = slightly acceptable, 7 = moderately acceptable, 8 = Very much acceptable and 9 = extremely acceptable. (Ranganna, 1994). All the samples were served in the Petri dishes and were returned for further chemical analysis.

(6) *Statistical analysis:* The analysis was carried out in triplicate and the mean values were compared using Duncan Post hoc tests, by SPSS program version 10.0 statistical software, at P value <0.05 to be significant.

3. Results and analysis

3.1. Effect of lactic acid concentration on the microbial count of fresh pork packed in MAP

The results revealed that the use of lactic acid spray reduced the microbial count and the effect was higher as the concentration of acid increased (Fig. 3). In each treatment there was a significant reduction of bacterial load with increasing acid concentration. On the other hand, the number of bacteria also increased significantly in all the treatments with increasing days of storage from 4th to 12th day. Microbial count on 4th day storage in the modified atmosphere was found to be 4.06 log cfu/gm for 6% lactic acid pretreated samples, while the value for the sample without lactic acid pretreatment was 1 log cycle more. The bacterial count was in the decreasing order from 1% lactic acid treated samples to 6% lactic acid treated samples. The maximum microbial load occurred in the control was 6.33 log cfu/gm on twelfth day's storage. On the other hand, higher lactic acid concentrations 4% and 6% treated samples exhibited 5.17 and 5.07 log cfu/gm, respectively on 12th day. The difference, though significant, of 1 and 2% lactic acid treated samples was relatively lower as compared to 2% and 4% lactic acid treated samples.

Regarding the microbial decontamination with lactic acid, the use of higher concentration exhibited better effect. This result seemed to be supported by the findings of Woolthuis and Smulders (1985). Smulders and Woolthus (1985) state that the hot-boned cuts of beef treated with 2% lactic acid spray could prolong the shelf life of the vacuum packaged beef. Beimuller et al. (1973) also report that 1–2 log of the total aerobic bacterial population can be reduced by the organic acid application on pork carcass. And recently in our result, the reduction of microbial load also took place with lactic acid treatment however it seemed relatively lower. What it assures is the destruction of the pathogens, if lactic acid decontamination is applied.

3.2. Effect of lactic acid concentration on the TBA values of fresh pork packed in MAP

The data showed that the TBARS values of the meat increased with the increasing days of storage and it existed also in the case of lactic acid treated samples (Fig. 4). With the increase in lactic acid concentration the TBARS values increased but significant difference existed only when the difference in acid concentrations was relatively higher such as in 1% and 4% or 2% and 6%. The lower concentration of acids did not increase the TBARS values significantly when compared with control treated with distilled water. Because of the high oxygen atmosphere, the initial TBA values on 4th day's storage analysis appeared relatively higher which were 0.276 and 0.372 mg malonaldehyde/kg, respectively for control and 6% lactic acid treated samples. Upto 2% of lactic acid application as compared with 0% Lactic acid, there was not a significant increment in the TBA values during 8th and 12th day's storage of fresh pork in the MAP. However, there was significant increment observed in TBA values for 4% and 6% Lactic acid treated samples.

Regarding the TBA values of meat, the use of lactic acid at lower concentration did not bring higher changes in the TBA values however the application at higher concentration of lactic acid increased the TBA value signifying the lipid oxidation. The higher amount of lactic acid might have denatured the surface protein releasing the iron (non-heme) which eventually facilitated the fats to oxidize (Erickson, 1998).

3.3. Effect of lactic acid concentration on the pH of the fresh meat packed in MAP

Normally the pH values of the meat did not change during storage of the fresh meat in MAP (Fig. 5). The lower concentration of the lactic acid spray such as at 1 and 2% did not bring larger change in pH while the higher concentration at 4 and 6% brought a significant change in pH. Such changes became more obvious

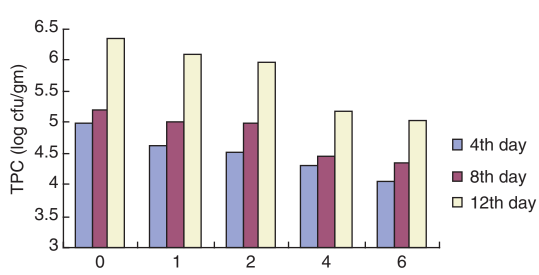


Fig. 3. Effect of lactic acid on the TPC value of fresh pork packed in MAP.

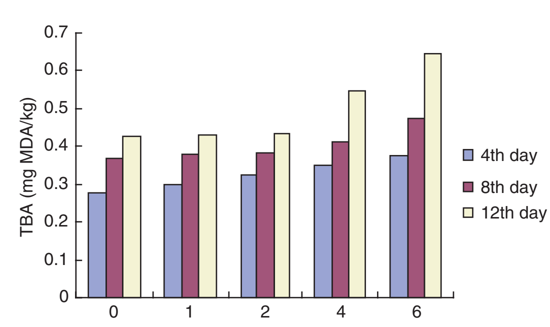


Fig. 4. Effect of lactic acid on the TBA values of fresh pork packed in MAP.

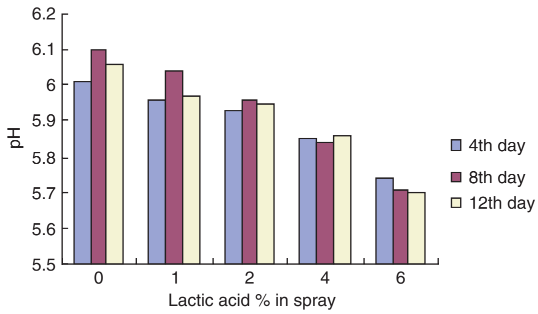


Fig. 5. Effect of lactic acid on the pH values of fresh pork packed in MAP.

during the later period of storage. For the higher content of lactic acid treated samples the pH values did not rise very high during storage. The pH values of the samples treated with lower level of lactic acid were found to have relatively higher pH than what normally required in meat storage.

Regarding the pH change in the meat, the use of lactic acid might have initially dropped the pH but the meat might have retained its pH value during storage like in the case stated by Woolthuis and Smulders (1985). The pH values decreased with the increasing concentration of the lactic acid (Van Netten, Huis In't Veld, & Mossel, 1994), and the similar style of decrement in pH was observed in this work. Greer and Dilts (1995) report that the surface pH of the lean pork treated with 3% lactic acid decreased in the beginning but after 24 h it rose and showed a little variation during the 12 days storage.

3.4. Effect of lactic acid concentration on the color scores of the fresh meat packed in MAP

The results revealed that the use of lactic acid spray reduced the color acceptability (1 for extremely dark brown and 9 for extremely bright pink) with the increasing concentration of lactic acid (Fig. 6). Generalizing the effect, the lower concentrations 1 and 2% did not reduce the color acceptability significantly when compared with the distilled water treated sample, stored for 4 days. In all those cases the scores ranged from 7.5 to 7.8. The higher concentrations 4 and 6% lactic acid reduced the color acceptability (color scores 7 to 6.8) and there was not significant difference observed between the treatments. In all the cases there was significant loss of color acceptability on 12th day storage (color scores ranged from 6.9 to 5.9). However, there was not such significant difference observed between 4th and 8th day's analysis.

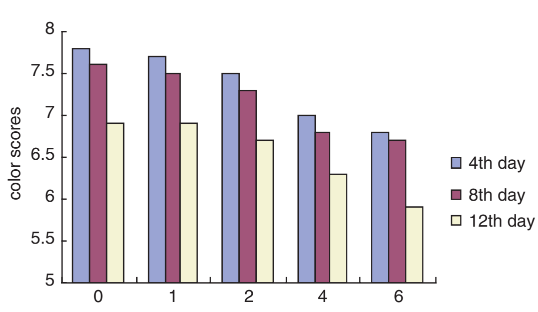


Fig. 6. Effect of lactic acid on the color scores of fresh pork packed in MAP.

Regarding the color scores of the lactic acid treated samples the acid had adverse effect on the color stability. In the lower lactic acid concentrations, the discoloration effect was lower, which was complying with the statement of Smulders (1987). It is stated that the 2% (v/v) lactic acid spray on deboned meat did not produce noticeable bleaching effect (Smulders & Woolthus, 1985). At the higher lactic acid concentrations, as observed in our work, the color appearance was significantly impaired. Such as 4 and 6% lactic acid treated samples could not attain the attractive fresh red color. Smulders (1987) states that the higher concentration of the lactic acid has bleaching effect on lean and discoloration effect on fats, which actually shows an agreement with the findings in this work.

3.5. Effect of lactic acid concentration on *a** values of the fresh meat packed in MAP

The data reveal that the *a** values of the samples pretreated with lactic acid and stored in MAP showed decrement with increasing days of storage (Fig. 7). Further, the use of lactic acid treatment had negative effect on *a** values exhibiting the significant loss of redness with increased lactic acid concentration.

The effect was more obvious at higher lactic acid concentrations such as 4 and 6%. On storage up to 12 days, the samples treated, even with lower concentration of lactic acid, exhibited significant loss in *a** values. The harsh effect of lactic acid on *a** value was not obvious on 4th day analysis but on 8th day it became more obvious for 4% and 6% lactic acid treated samples (the values were 7.37 and 6.81, respectively). However, on 12th day analysis, the differences in *a** values among

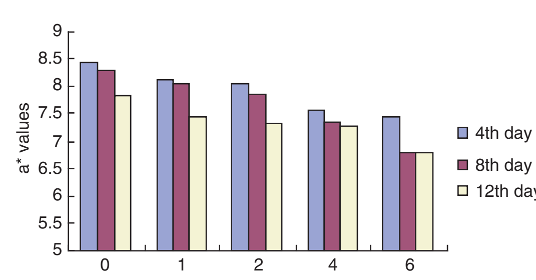


Fig. 7. Effect of lactic acid on *a** value of fresh pork packed in MAP.

the samples were relatively lower, as the samples which were better in the earlier period also had undergone deterioration.

Regarding the instrumental color parameter 'a^* value', our result seemed supported with the statement of Arganosa and Marriott (1989) in which a^* values were observed to be lower for lower pH meat, treated with lactic acid. Redness of the meat is represented by a^* values (Van Laack, Berry, & Solomon, 1996), and hence the loss of redness shows decrease in a^* values (Young & West, 2001) and vice-versa. The trend of a^* value decrement seemed to be positively correlated with sensory color scores, significant at 0.01 level (Pearson's correlation coefficient 0.919).

3.6. Effect of lactic acid concentration on the overall acceptability scores of the fresh meat packed in MAP

The overall acceptability (1 for extremely unacceptable and 9 for extremely acceptable) of the lactic acid treated samples decreased with the increasing concentration of the lactic acid in the spray as seen in Fig. 8. The control sample treated with distilled water alone has the maximum acceptability on 4th day's storage exhibiting the overall acceptability scores of 7.8 while that of 6% lactic acid treated sample was 6.83. On 12th day the loss of the overall acceptability was very obvious in all the treatments. There was no significant difference observed for 0, 1, 2 and 4% lactic acid treated samples where 0% and 4% had scores respectively 6.83 and 6.16. Further, there was a little effect observed for all the treatments between 4 and 8 days storages.

The overall acceptability was evaluated in terms of the color, odor, drip loss, and overall appearance of the meat. There was decrease in scores with higher drip loss, dry surface, poor color and bad odor. Thus, which particular term was more important could not be specified in this overall score values. The color scores and the overall acceptability scores seemed to be highly correlated signifying that, color score has a significant influence on the overall acceptability scores.

3.7. Effect of lactic acid pretreatment on the lightness L^* value of the fresh pork packed in MAP

The results revealed that the lightness L^* values of the fresh meat packed in MAP containing 45% O_2 and 20% CO_2 decreased with the increasing period of storage (Fig. 9). The application of lactic acid pretreatment exhibited a decrement in the L^* values in the early 4 days storage according to the increase in the acid concentration. The deionized water added sample had relatively higher L^* value 39.0 while that of 6% lactic acid treated sample was 36.78. During the storage the decrement in L^* value of the 0% Lactic acid treated sample was found to be the highest among the treatments. There were significant decrements in 0,1and 2% Lactic acid treated samples stored between 4 and 12 days. For other treatments there was not such significant decrement observed during storage though the trend seems to be in decreasing order.

The lightness L^* values for the pork observed relatively lower in our experiment indicates the meat tending towards DFD. The lightness value of the sample depends upon the moist surface which has higher reflecting property (Young & West, 2001). During the chilled storage, the surface of meat might have dried a bit due to evaporation.

3.8. Effect of lactic acid concentration on Hue angle of the fresh meat packed in MAP

Fig. 10 shows that there was an increment of the hue angle with the storage period of the fresh pork treated with lactic acid and packed in MAP. It also indicates that the hue angle given by $\tan^{-1}(b^*/a^*)$, increased with the increasing concentration of the lactic acid in the treatment solutions.

For the 4th day analysis there was no significant increment of the hue angle observed with the increasing concentration of acid. However, on 8th and 12th days, only the 6% LA treated sample differed significantly from 0, 1 and 2% LA treated samples.

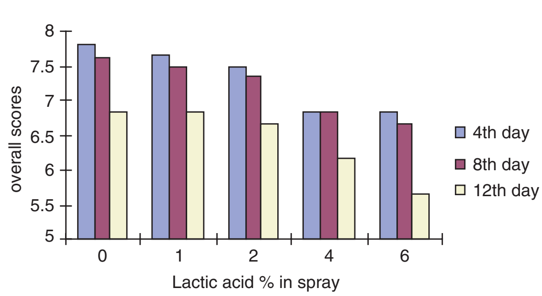


Fig. 8. Effect of lactic acid on overall acceptability of fresh pork packed in MAP.

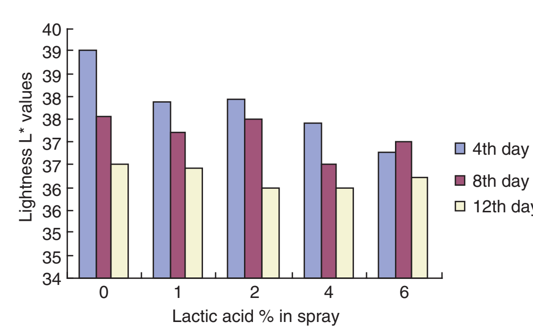


Fig. 9. Effect of lactic acid on lightness L^* values of fresh pork packed in MAP.

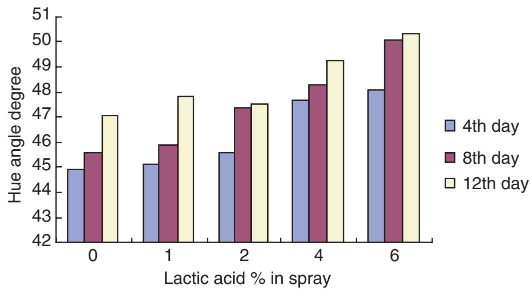


Fig. 10. Effect of lactic acid on Hue angle of fresh pork packed in MAP.

The increase or decrease in the hue angle depends upon change of a^* and b* values. For the increase of the 'a^*' value without change in b* value, the hue angle decreases and the reverse is true. Increment of hue angle shows the decrement of redness (Van Laack et al., 1996). From the previous results, there was a decrement of redness with the increasing concentration of lactic acid in the treatment solutions; hence the results are well correlated.

4. Conclusions

From the above results and discussions it can be concluded that lactic acid application in the fresh meat could reduce the bacterial load, but the better effects could be observed only at 4 and 6% lactic acid concentrations (1 log reduction in bacterial count). However, there was a significant loss of color and overall acceptability scores along with significant increment in the TBA values. Therefore, to enhance the effect of lactic acid application in fresh meat packed in MAP, suitable color and lipid stabilizers need to be sought.

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