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MICROBIAL ASPECTS OF LAMB MEAT TREATED WITH LACTIC AND ACETIC ACIDS (With 8 Tables)

By

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الوجهة الميكروبية للحوم الضأن المعاملة بحمض اللاكتيك والخليك

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أجريت هذه الدراسة لتقييم قدرة الأحماض العضوية على تحسين جودة لحوم الضأن الطازجة نوعيا عند ٢٤ و ٤٨ و ٧٢ ساعة و ٧ و ١٤ يوم بتطبيق عملية التبريد بعد معالجتها بالأحماض العضوية (حمض اللاكتيك ١% و ٢% حمض الخليك ١% و ٢%) نتج عن هذه المعالجة لعدد ٢٥ عينة قيد الدراسة والتي أخذت منها بعض الأجزاء ككاشف تأثيرات معنوية ضد البكتريا كان لها الأثر الأكبر في أطاله عمر وصلاحية اللحوم المبردة - أظهر قياس درجة الحموضة ارتفاعات بلغت أقصاها عند التبريد لمدة ١٤,٧ يوم بعد المعاملة - أحدثت المعاملة بالأحماض العضوية تأثيرات دراماتيكية على المحتوى الميكروبي للحوم الضأن المعاملة مقارنة بالعينات التي تركت ككاشف. حيث أوضح العدد الكلي للميكروبات الهوائية انخفاضا معنويا تتناسب طرديا مع فترة التبريد حيث بلغ الاختزال في متوسطات العدد البكتيري أقصاه عند اليوم السابع للتبريد ووصلت ٧٠,٨٧ , ٦٤,٠٩ , ٧١ و ٦٠,٤٥% بالمعاملة بـ ١% و ٢% حمض لكتيك و ١% و ٢% حمض الخليك على التوالي كذلك وصل الاختزال في حالة التبريد تحت الصفر ٦٢,٣٧ و ٦٢,٥٣ و ٥٧٩٦ و ٥٧,٣٨% في ذات المعاملات على التوالي كذلك بالنسبة للعدد الكلي للميكروبات المحية للبرودة فقد بلغ الاختزال بينهما أقصاه عند اليوم السابع حيث كانت ٤٩,٥٤ و ٤٣,٣٣ و ٥٣,٣٦ و ٣٨,٠٤% لذات المعاملات بالتوالي. كذلك الاختزال للمعاملة عند التبريد تحت الصفر بلغت ٢٦,٤١ و ١٥ و ٢٦,٧١ و ١٥,٨٨% أحدثت المعاملة بالأحماض خفض في المحتوى للعدد الكلي للميكروبات القولونية بلغ أقصاه في اليوم السابع وبلغت نسبة الاختزال فيها ٦٨,٠٦ و ٦٥,٢٤ و ٧١,٨١ و ٦٤,٧٧% على التوالي - كانت نسبة الاختزال في التبريد تحت الصفر ٣٥,٠٦ و ٥,٧٢ و ٥٥,١٢ و ٤٨,٢٨% - أدت المعاملات لاختزال العدد البكتيري للسالمونيلا حيث كانت في العينات الطازجة ٤,٨ × ١٠ + ٣,٢ × ١٠^٢ ولكن بعد المعاملة وصلت إلى الصفر عند اليوم السابع وكذلك عند التجميد - العدد البكتيري للمكورات العنقودية انخفض بدرجة كبيرة بعد المعاملة ووصلت نسبة الاختزال في المتوسطات في اليوم السابع إلى ٤٢,٨٦ و ٦٠,٥٣ و ٥٩,٤٦ و ٣٣,٣٣% بينما عند التجميد وصل الاختزال إلى ٧,١٤ و ٦٤,٩١ و ٥٥,٤١ و ٦٠% على التوالي - العدد الكلي للفطريات والخمائر أظهر انخفاضا شديدا بعد

المعاملة بالأحماض حيث وصلت بعد ٧٢ ساعة إلى حدود دنيا وبلغ عندها نسبة الاختزال ٥٦,٧٤ و ٥٢,٠٧ و ٥٢,٩٤ و ٣٩,٠٢% بينما في مرحلة التجميد حدث ارتفاع بسيط في المتوسطات بالرغم من انخفاض نسبة الاختزال عما كان متوقع في هذه المرحلة - كان للمعاملة بالأحماض العضوية أثرا معنويا على البكتيريا المحللة للبروتين حيث وصلت نسبة الاختزال في العد البكتريا لها ١٣,٤٩ و ٤٢,٢٦ و ٢٢,٩٩ و ٣٨,٦٥% عند المعاملة بحمض اللاكتيك ١% و ٢% وحمض الخليك ١% و ٢% على التوالي أما البكتيريا المحللة للدهون فكانت نسبة الاختزال في العدد ٣٢,٨٦ و ٥٩,٨٧ و ٣١,٨٦ و ٦٠,٨٨% في ذات المعاملات على التوالي تم تصنيف العصيات القولونية المعزولة عبر مراحل الدراسة واتضح التأثير بالمعاملة الحمضية حيث ٢% أحدثت انخفاضا أكثر من ١% دون تأثير المواصفات الطبيعية نفس النتيجة لوحظت في السالمونيلا الملهبية والمكورات العنقودية الذهبية والفطريات المختلفة هذا وقد تم مناقشة الأهمية الصحية والاقتصادية للميكروبات المعزولة وشرح أهمية استخدام الأحماض العضوية (الخليك واللاكتيك) في خفضها واثار ذلك على الصحة العامة

SUMMARY

The present study was carried out to assess the ability of organic acids to improve the quality of fresh lamb meat chilled at 7 °C for 24, 48, 72 hours, 7 & 14 day and frozen at -18 °C after treatment with organic acids (lactic and acetic 1 & 2%) which resulted in significant improvement of nearly all parameters over the control of 25 samples from different lamb carcasses. Such treatments exerted a significant antibacterial effect which is of public health importance and prolong the shelf life of carcasses on trial. The pH value revealed highest record at 14 day chilling. The organic acids revealed dramatic effects as antimicrobial agent. The total aerobic bacterial count revealed reciprocal reduction percent 70.87, 64.09, 71.07 and 60.54% in chilling for 7 day after treatment with 1 & 2% lactic and acetic acid respectively. The reciprocal reduction percents were 62.37 to 62.53% and 57.96 and 57.38% in 1 & 2% lactic and acetic acids. On applying the psychrophilic bacteria the reciprocal reduction percent means at the 7th day of chilling were 49.54, 73.33, 53.36 and 38.04% respectively with same acids treatments. On apply freezing for one month the reduction values reciprocal to the means were 29.41, 15.00, 26.71 and 15.88% respectively. The lowest records of total coliform counts were shown on the 7th day chilling. The reciprocal reduction percent of means were 73.65, 65.04, 77.84 and 68.59. After month of freezing the corresponding reductions were 53.51, 45.51, 63.72 and 45% with acid treatments respectively. The reduction percent of total Enterobacteriaceae counts of the means were 68.06, 62.24, 71.81 and 64.77% after 7 days of chilling lamb meat post acid

treatment. Salmonella count revealed means zero after 7, 14 day of chilling lamb meat; also, means were zero after freezing for month post acids treatments. Staphylococcus aureus count: revealed highest reduction at the 7th day post chilling and the reciprocal reductions were 42.86, 60.53, 59.46 and 33.33% respectively. Total mould and yeast count showed reduction percents of 56.74, 52.07, 52.94 and 39.02% after 72 hour of treated chilled lamb meat. After freezing, reduction percents were 26.97, 29.75, 16.99% in lactic acid 1, 2% and acetic acid 1% where count increased in acetic acid 2%. The means of Proteolytic bacterial count reduced to 13.49, 42.26, 22.99 and 38.65% in 1, 2% lactic acid and 1 & 2% acetic acid treated lamb carcasses. The lipolytic bacterial count reductions were 32.86, 59.37, 31.86 and 60.88% respectively. Coliforms were detected in lower incidence in the treated sample including *E.coli*, *Enterobacter* spp, *Klebsiella* spp., *Edwardesiella tarda* and *Serratia rubidea*. The acid treatment lower the occurrence of coliform where 2% concentration was more efficient than 1%. The same results were detected in the identified *Staphylococcus aureus* and *Salmonella enteritidis* in addition to the identified moulds

Key words: *Organic acids, lactic acid, acetic acid, lamb carcasses.*

INTRODUCTION

Meat is an important vehicle for food borne diseases such as salmonellosis and campylo-bacteriosis. Many other organisms act as toxin producers and spoilage formers. Organic acids including acetic, fumaric, propionic, and lactic acids are added to foods to prevent or delay the growth of pathogenic or spoilage bacteria. The inhibitory effects of acids on microbial growth has long been used to preserve foods from spoilage (Podolak, *et al.*, 1996).

The external contamination of meat constitute a constant problem in meat developing countries where the abattoirs itself have a large numbers of potential sources of contamination (Davis *et al.*, 2000). Organic acids as antimicrobial agents for surface treatment of fresh meat have been used to prevent the growth bacteria during chill storage (ICMSF, 1982). Similarly in Europe it is considered a harmless constituent (leuck, 1980). This widely knowledge about absence of acute and chronic toxicity has led to the choice of lactic acid as decontaminating agent in food industry. Data are available on the potency of lactic acid spray as carcass decontaminant for lamb and beef

(Fatema-Ali, 2001) Lactic and acetic acids are generally recognized as safe food additives. Both acids are based as apart of meat decontamination procedures, where they are more effective than many other technique and components (Gorman, *et al.*, 1997). Numerous studies have reported on the effects of organic acids on bacterial populations as well as some pathogenic organisms (Dickson and Anderson, 1991, Hardin, *et al.*, 1995 and Castillo, *et al.*, 2001).

The object of the current research was to evaluate the effectiveness of acetic and lactic acids on microbial loads and characteristics of treated lamb carcasses across the chilling and freezing time.

MATERIALS and METHODS

Collection of samples: From a total of 25 fresh representative lamb carcasses, randomly collected from different butchers shops at Behaira province, right fore limb and left hind limb were taken and packed in insulating containers and transferred as soon as possible to the laboratory for microbiological examination

Treatment of samples: The fresh samples without treatment were taken as control. Samples were sprayed with aqueous solutions of lactic acid 1 and 2%, others were sprayed with acetic acid 1 and 2% and all samples were hanged for ten minutes to dry. Samples were put in chilling at 7 °C for 24, 48, 72 hours, 7 and 14 days and frozen at -18 °C for one month.

Preparation of samples: Was done according to ICMSF (1982)

Analysis:

- Measurement of PH was determined by using Digital pH. Meter
- Microbial Examination
 - Determination of Total aerobic bacterial count according to ICMSF (1982)
 - Determination of Psychrophilic bacterial count according to ICMSF (1982)
 - Determinatio of Total Enterobacteriaceae count according to ICMSF (1978).
 - Estimation of coliform according to ICMSF (1982)
 - Estimation and detection of Salmonellae: was carried according to Andrews and AOAC (1984) Biochemical identification of Enterobacteriaceae and Salmonellae were carried according to Cruickshank, *et al.* (1975) and ICMSF (1982)

- Determination of staphylococci was done according to Cruickshank, *et al.* (1975) and ICMSF (1978)
- Biochemical identification of *Staphylococcus aureus* was carried according to (APHA, 1984)
- Total mould and yeast count was carried according to Bailey and Scott, (1978)
- Identification of moulds was carried according to Raper and Fennel (1965) and Samson, *et al.* (1995) for genes *Aspergillus* and *Penicillium*, while other genera were identified according to Zycha, *et al.* (1969), Barnett and Hanter (1972) and Samson, *et al.* (1995)
- Identification of yeasts was carried according to Lodder (1967)

RESULTS

Table 1: Means of pH of the examined treated lamb carcasses stored at 7 °C

Treatments Duration	Treatment (means ± standard errors)			
	Lactic acid 1 %	Lactic acid 2 %	Acetic acid 1 %	Acetic acid 2 %
24 hr chilling	3.26 ± 0.05 a	3.06 ± 0.02 b	3.36 ± 0.02 a	3.06 ± 0.02 b
48 hr chilling	0.50 ± 0.03 ab	3.26 ± 0.02 c	3.58 ± 0.02 a	3.32 ± 0.04 ab
72 hr chilling	4.06 ± 0.02 a	3.54 ± 0.02 b	4.14 ± 0.05 a	3.064 ± 0.07 b
7days chilling	5.46 ± 0.05 a	4.86 ± 0.22 c	5.52 ± 0.09 a	5.10 ± 0.05 b
14 days chilling	6.08 ± 0.04 a	5.60 ± 0.08 b	6.10 ± 0.06 a	5.72 ± 0.4 b

Means in the same row followed by a similar letter do not differ significantly at $p=0.05$.
PH of control samples = 5.60 ± 0.03 .

Table 2: Means of total aerobic and total psychrophilic bacterial counts (cfu / g) of the examined lamb carcasses before and after application of acid treatment (n = 25)

		Treatments (Means \pm standard errors)							
		Lactic acid 1%		Lactic acid 2%		Acetic acid 1%		Acetic acid 2%	
Chilling		Total aerobic	Total psychr	Total aerobic	Total psychr	Total aerobic	Total psychr	Total aerobic	Total psychr
	Fresh	2.14 $\times 10^3 \pm$ 2.25 $\times 10^2$ a	1.25 $\times 10^3 \pm$ 1.67 $\times 10^2$ a	1.54 $\times 10^3 \pm$ 2 $\times 10^2$ bc	1.2 $\times 10^3 \pm$ 1.5 $\times 10^2$ ab	2.06 $\times 10^3 \pm$ 2.44 $\times 10^2$ ab	1.46 $\times 10^3 \pm$ 1.82 $\times 10^2$ c	1.3 $\times 10^3 \pm$ 1.89 $\times 10^2$ c	1.02 $\times 10^3 \pm$ 1.21 $\times 10^2$ b
	24 hr	1.37 $\times 10^3 \pm$ 1.31 $\times 10^2$ a	1.26 $\times 10^3 \pm$ 1.02 $\times 10^2$ a	8.8 $\times 10^2 \pm$ 1 $\times 10^2$ b	1.03 $\times 10^3 \pm$ 0.92 $\times 10^2$ a	1.42 $\times 10^3 \pm$ 1.34 $\times 10^2$ a	1.11 $\times 10^2 \pm$ 0.86 $\times 10^2$ a	8.56 $\times 10^2 \pm$ 1.08 $\times 10^2$ b	9.9 $\times 10^2 \pm$ 1.2 $\times 10^2$ b
	48 hr	1.01 $\times 10^3$ 1 $\times 10^2$ a	9.34 $\times 10^2 \pm$ 0.74 $\times 10^2$ a	7.26 $\times 10^2 \pm$ 0.93 $\times 10^2$ b	7.06 $\times 10^2 \pm$ 0.7 $\times 10^2$ a	1.03 $\times 10^3 \pm$ 1.31 $\times 10^2$ a	9.2 $\times 10^2 \pm$ 0.82 $\times 10^2$ a	7.15 $\times 10^2 \pm$ 1.08 $\times 10^2$ b	7.02 $\times 10^2 \pm$ 0.75 $\times 10^2$ a
	72 hr	1.13 $\times 10^3 \pm$ 2.05 $\times 10^2$ a	8.55 $\times 10^2 \pm$ 0.84 $\times 10^2$ a	5.99 $\times 10^2 \pm$ 0.75 $\times 10^2$ bc	6.56 $\times 10^2 \pm$ 0.77 $\times 10^2$ b	7.92 $\times 10^2 \pm$ 0.92 $\times 10^2$ ab	7.8 $\times 10^2 \pm$ 0.89 $\times 10^2$ a	5.57 $\times 10^2 \pm$ 0.76 $\times 10^2$ c	6.36 $\times 10^2 \pm$ 0.77 $\times 10^2$ b
	7 Days	6.38 $\times 10^2 \pm$ 0.53 $\times 10^2$ a	7.72 $\times 10^2 \pm$ 0.65 $\times 10^2$ a	5.53 $\times 10^2 \pm$ 0.51 $\times 10^2$ a	6.8 $\times 10^2 \pm$ 0.66 $\times 10^2$ a	5.96 $\times 10^2 \pm$ 0.51 $\times 10^2$ a	6.81 $\times 10^2 \pm$ 0.72 $\times 10^2$ a	5.13 $\times 10^2 \pm$ 0.5 $\times 10^2$ a	6.32 $\times 10^2 \pm$ 0.64 $\times 10^2$ a
	14 Days	9.62 $\times 10^2 \pm$ 1.08 $\times 10^2$ a	1.02 $\times 10^2 \pm$ 0.59 $\times 10^2$ a	9.07 $\times 10^2 \pm$ 0.61 $\times 10^2$ a	9.21 $\times 10^2 \pm$ 0.95 $\times 10^2$ a	8.85 $\times 10^2 \pm$ 0.77 $\times 10^2$ a	9.16 $\times 10^2 \pm$ 0.68 $\times 10^2$ a	7.89 $\times 10^2 \pm$ 0.72 $\times 10^2$ a	8.62 $\times 10^2 \pm$ 0.95 $\times 10^2$ a
	Freezing	8.24 $\times 10^2 \pm$ 1.08 $\times 10^2$ a	1.08 $\times 10^2 \pm$ 1.7 $\times 10^2$ a	5.77 $\times 10^2 \pm$ 0.94 $\times 10^2$ a	1.02 $\times 10^3 \pm$ 1.3 $\times 10^2$ a	6.66 $\times 10^2 \pm$ 1.2 $\times 10^2$ a	1.07 $\times 10^2 \pm$ 1.44 $\times 10^2$ a	5.54 $\times 10^2 \pm$ 0.95 $\times 10^2$ b	8.52 $\times 10^2 \pm$ 1.38 $\times 10^2$ a

Means in the same raw followed by same letters do not differ significantly at $p=0.05$ within each of aerobic and Psychrophilic counts

Mean value of control samples in total aerobic bacterial count = $3.74 \times 10^3 \pm 2.88 \times 10^2$

Mean value of control samples in total Psychrophilic bacterial count = $2.06 \times 10^3 \pm 2.85 \times 10^2$

Table 3: Reduction percentages of total aerobic, psychrophilic and *Staph. aureus* count of the examined treated lamb carcasses (n=25)

Treat	Lactic acid 1%			Lactic acid 2%			Acitic acid 1%			Acitic acid 2%		
	Aerobic	psychro.	Staph.	aerobic	psychro.	Staph.	aerobic	psychro.	Staph.	aerobic	psychro.	Staph.
Fresh	41.44	25.73	42.62	58.82	41.75	53.28	44.14	92.24	39.34	65.24	50.49	69.26
24 h. Chil	37.44	17.65	+22.14*	42.86	14.17	+10.53*	31.07	23.97	+15.54*	34.15	2.75	+60.00*
48 h. Chil	53.88	38.59	2.868	52.86	41.17	40.35	50.00	38.63	0.00	45.00	31.18	+13.33
72 h. Chil	48.40	44.12	37.14	61.10	45.33	3.51	61.55	46.58	29.05	57.15	37.65	+6.67*
7 d. Chil	70.87	49.45	42.86	64.09	43.33	60.53	71.07	53.36	59.46	60.54	38.04	33.33
14 d. Chil	57.70	33.33	7.14	41.10	23.25	29.28	57.04	37.26	48.00	39.31	15.49	2.67
Freezing	62.37	29.4	7.14	62.53	15.00	64.91	57.96	26.71	55.41	57.38	15.88	60.00

The reduction percent was the deviation of treated means relative to control mean in the fresh state in chilling and freezing state
 (It was the deviation of treated means relative to the treatment means in fresh state)

Chil = Chilling aerob. = aerobic Psychro = Psychrophilic

Table 4: Statistical analytical results of means of total coliform (M.P.N/g) & Enterobacteriaceae count (cfu/g) of examined lamb carcasses before and after application of acid treatment (n = 25)

		Treatments (Means \pm standard errors)							
		Lactic acid 1%		Lactic acid 2%		Acetic acid 1%		Acetic acid 2%	
		coliform	enterob	Coliform	Enterob	coliform	enterob	coliform	enterob
Fresh		$9.83 \times 10^2 \pm$ 1.31×10^2 a	$1.24 \times 10^3 \pm$ 1.7×10^2 a	$6.46 \times 10^2 \pm$ 0.99×10^2 b	$9.33 \times 10^2 \pm$ 1.59×10^2 ab	$1.02 \times 10^2 \pm$ 1.41×10^2 a	$1.27 \times 10^2 \pm$ 1.94×10^2 a	$6.4 \times 10^2 \pm$ 0.84×10^2 b	$3.43 \times 10^2 \pm$ 1.34×10^2 b
Chilling	24 hr	$6.37 \times 10^2 \pm$ 0.91×10^2 a	$9.55 \times 10^2 \pm$ 1.55×10^2 a	$4.54 \times 10^2 \pm$ 0.77×10^2 a	$6.36 \times 10^2 \pm$ 1×10^2 b	$6.31 \times 10^2 \pm$ 0.86×10^2 a	$9.72 \times 10^2 \pm$ 1.69×10^2 b	$4.47 \times 10^2 \pm$ 0.68×10^2 a	$6.33 \times 10^2 \pm$ 1.02×10^2 b
	48 hr	$5.02 \times 10^2 \pm$ 0.87×10^2 a	$6.79 \times 10^2 \pm$ 0.87×10^2 a	$4.02 \times 10^2 \pm$ 0.87×10^2 a	$4.56 \times 10^2 \pm$ 0.71×10^2 bc	$5.21 \times 10^3 \pm$ 0.99×10^2 a	$6.5 \times 10^2 \pm$ 0.94×10^2 ab	$3.83 \times 10^2 \pm$ 0.85×10^2 a	$4.2 \times 10^2 \pm$ 0.65×10^2 c
	72 hr	$3.86 \times 10^2 \pm$ 0.65×10^2 a	$5.47 \times 10^2 \pm$ 0.73×10^2 a	$3.06 \times 10^2 \pm$ 0.58×10^2 a	$3.74 \times 10^2 \pm$ 0.65×10^2 b	$3.89 \times 10^2 \pm$ 0.66×10^2 b	$5.46 \times 10^2 \pm$ 0.87×10^2 ab	$2.97 \times 10^2 \pm$ 0.63×10^2 a	$3.6 \times 10^2 \pm$ 0.64×10^2 b
	7 Days	$2.95 \times 10^2 \pm$ 0.37×10^2 a	$3.69 \times 10^2 \pm$ 0.43×10^2 a	$2 \times 10^2 \pm$ 0.34×10^2 a	$3.25 \times 10^2 \pm$ 0.4×10^2 a	$2.26 \times 10^2 \pm$ 0.35×10^2 a	$3.58 \times 10^2 \pm$ 0.4×10^2 a	$2.01 \times 10^2 \pm$ 0.34×10^2 a	$2.97 \times 10^2 \pm$ 0.38×10^2 a
	14 Days	$4.63 \times 10^2 \pm$ 0.48×10^2 a	$5.86 \times 10^2 \pm$ 0.47×10^2 a	$3.93 \times 10^2 \pm$ 0.43×10^2 a	$4.9 \times 10^2 \pm$ 0.45×10^2 a	$4.34 \times 10^2 \pm$ 0.43×10^2 a	$5.34 \times 10^2 \pm$ 0.47×10^2 b	$3.46 \times 10^2 \pm$ 0.43×10^2 a	$4.41 \times 10^2 \pm$ 0.45×10^2 a
Freezing		$4.57 \times 10^2 \pm$ 1.14×10^2 a	$5.82 \times 10^2 \pm$ 1.28×10^2 a	$3.52 \times 10^2 \pm$ 0.94×10^2 a	$4.14 \times 10^2 \pm$ 1.18×10^2 a	$3.7 \times 10^2 \pm$ 0.8×10^2 a	$5.7 \times 10^2 \pm$ 1.35×10^2 a	$3.52 \times 10^3 \pm$ 0.94×10^2 b	$4.36 \times 10^2 \pm$ 1.31×10^2 a

Enterob = Enterobacteriaceae

Means in the same raw followed by same letters do not differ significantly at $p=0.05$ within each of Coliform and Enterobacteriaceae countMean value of control samples in total Coliform count = $1.53 \times 10^3 \pm 1.82 \times 10^2$ Mean value of control samples in total Enterobacteriaceae count = $2.00 \times 10^3 \pm 2.55 \times 10^2$

Table 5: Reduction percentages of total coliform, Enterobacteriaceae, *Salmonellae* and Mould & Yeast count of examined treated lamb carcasses.

Treat.	Lactic acid 1%				Lactic acid 2%				Acetic acid 1%				Acetic acid 2%			
	Coli.	Ent.	Salm.	Yeast & Mold	Coli.	Ent.	Salm.	Yeast Mold	Coli.	Ent.	Salm.	Yeast Mold	Coli.	Ent.	Salm.	Yeast Mold
Fresh	35.75	38.00	45.83	39.25	57.78	53.25	75.0	58.70	33.33	36.50	41.14	47.78	58.14	57.85	72.92	72.01
24 h	35.20	22.98	56.54	30.90	29.72	32.41	41.67	42.98	38.14	23.46	50.0	20.92	30.16	24.91	53.85	17.07
48 h	48.94	45.24	65.38	42.13	37.77	51.23	60.83	47.93	48.92	64.29	39.94	32.03	39.94	49.94	69.23	34.15
72 h	60.73	55.89	76.37	56.74	60.00	83.33	61.86	52.07	57.01	78.01	78.57	52.94	53.59	57.3	69.23	39.02
7 d	73.65	68.06	-	33.71	69.04	65.24	-	46.28	77.84	71.81	-	48.37	68.59	64.77	-	+8.54
14 d	52.90	52.74	-	16.83	39.16	47.59	-	17.36	57.49	57.95	-	19.61	45.94	47.69	-	8.54
Freezing	53.51	53.06	-	26.97	45.51	55.72	-	29.75	63.72	55.12	-	16.99	45.00	48.28	-	+3.66

The reduction percent was the deviation of treated means relative to control mean in the fresh state in chilling and freezing state

It was the deviation of treated means relative to the treatment means in fresh state

Coli=Coliform

Ent. = Enterobacteriaceae

Salm. = *Salmonellae*

Table 6: Statistical analytical results of means of total *Salmonellae* count, *Staphylococcus aureus* and Mould & yeast count (cfu/g) of the examined lamb carcasses before and after application of acid treatment (n = 25)

Treatments (Means \pm standard errors)													
		Lactic acid 1%			Lactic acid 2%			Acetic acid 1%			Acetic acid 2%		
		Total Salm.	Total Staph.	Mold & yeast	Total Salm.	Total Staph.	Mold & yeast	Total Salm.	Total Staph.	Mold & yeast	Total Salm.	Total Staph.	Mold & yeast
Chilling	Fresh	$2.6 \times 10^2 \pm 1.6 \times 10^2$ a	$1.4 \times 10^2 \pm 0.43 \times 10^2$ b	$1.78 \times 10^2 \pm 0.2 \times 10^2$ a	$1.2 \times 10^2 \pm 1 \times 10^2$ a	$1.14 \times 10^2 \pm 0.37 \times 10^2$ b	$1.21 \times 10^2 \pm 0.2 \times 10^2$ b	$2.8 \times 10^2 \pm 1.8 \times 10^2$ a	$1.48 \times 10^2 \pm 0.42 \times 10^2$ b	$1.53 \times 10^2 \pm 0.17 \times 10^2$ a	$1.3 \times 10^2 \pm 1.1 \times 10^2$ a	$0.75 \times 10^2 \pm 0.24 \times 10^2$ b	$0.73 \times 10^2 \pm 0.0.12 \times 10^2$ b
	24 hr	$1.13 \times 10^2 \pm 0.52 \times 10^2$ a	$1.71 \times 10^2 \pm 0.56 \times 10^2$ a	$1.23 \times 10^2 \pm 0.14 \times 10^2$ a	$0.7 \times 10^2 \pm 0.1 \times 10^2$ a	$1.26 \times 10^2 \pm 0.57 \times 10^2$ b	$1.26 \times 10^2 \pm 0.57 \times 10^2$ b	$1.4 \times 10^2 \pm 0.4 \times 10^2$ a	$1.71 \times 10^2 \pm 0.62 \times 10^2$ a	$1.21 \times 10^2 \pm 0.13 \times 10^2$ a	0.6×10^2 a	$1.2 \times 10^2 \pm 0.55 \times 10^2$ a	$0.68 \times 10^2 \pm 0.09 \times 10^2$ b
	48 hr	$0.9 \times 10^2 \pm 0.1 \times 10^2$ a	$1.36 \times 10^2 \pm 0.43 \times 10^2$ a	$1.03 \times 10^2 \pm 0.1 \times 10^2$ a	$0.47 \times 10^2 \pm 0.13 \times 10^2$ a	$0.68 \times 10^2 \pm 0.25 \times 10^2$ b	$0.63 \times 10^2 \pm 0.09 \times 10^2$ b	$1 \times 10^2 \pm 0.2 \times 10^2$ a	$1.48 \times 10^2 \pm 0.59 \times 10^2$ a	$1.04 \times 10^2 \pm 0.07 \times 10^2$ a	0.4×10^2 a	$0.85 \times 10^2 \pm 0.3 \times 10^2$ a	$0.54 \times 10^2 \pm 0.61 \times 10^2$ b
	72 hr	$0.6 \times 10^2 \pm 0.00$	$0.88 \times 10^2 \pm 0.34 \times 10^2$ a	$0.77 \times 10^2 \pm 0.08 \times 10^2$ a	$0.2 \times 10^2 \pm 0.00$ a	$1.1 \times 10^2 \pm 0.7 \times 10^2$ a	$0.58 \times 10^2 \pm 0.11 \times 10^2$ a	$0.6 \times 10^2 \pm 0.2 \times 10^2$ a	$1.05 \times 10^2 \pm 0.4 \times 10^2$ a	$0.72 \times 10^2 \pm 0.07 \times 10^2$ a	$0.4 \times 10^2 \pm 0.00$ a	$0.8 \times 10^2 \pm 0.6 \times 10^2$ a	$0.5 \times 10^2 \pm 0.06 \times 10^2$ a
	7 Day	--	$0.8 \times 10^2 \pm 0.22 \times 10^2$ a	$1.18 \times 10^2 \pm 0.16 \times 10^2$ a	--	$0.45 \times 10^2 \pm 0.19 \times 10^2$ b	$0.65 \times 10^2 \pm 0.09 \times 10^2$ b	--	$0.6 \times 10^2 \pm 0.22 \times 10^2$ a	$0.79 \times 10^2 \pm 0.09 \times 10^2$ b	--	$0.5 \times 10^2 \pm 0.3 \times 10^2$ a	$0.89 \times 10^2 \pm 0.36 \times 10^2$ b
	14 Day	--	$1.3 \times 10^2 \pm 0.3 \times 10^2$ a	$1.48 \times 10^2 \pm 0.08 \times 10^2$ a	--	$0.8 \times 10^2 \pm 0.25 \times 10^2$ bc	$1.0 \times 10^2 \pm 0.08 \times 10^2$ bc	--	$1 \times 10^2 \pm 0.45 \times 10^2$ a	$1.23 \times 10^2 \pm 0.08 \times 10^2$ ab	--	$0.73 \times 10^2 \pm 0.27 \times 10^2$ a	$0.75 \times 10^2 \pm 0.0.08 \times 10^2$ c
	Freez	--	$1.3 \times 10^2 \pm 0.7 \times 10^2$ a	$1.3 \times 10^2 \pm 0.11 \times 10^2$ a	--	0.4×10^2 b	$0.85 \times 10^2 \pm 0.08 \times 10^2$ b	--	$0.66 \times 10^2 \pm 0.47 \times 10^2$ a	$1.27 \times 10^2 \pm 0.1 \times 10^2$ a	--	1.2×10^2 a	$0.85 \times 10^2 \pm 0.0.09 \times 10^2$ b

Salm.=*Salmonella*Staph.= *Staphylococcus aureus*Means in the same raw followed by same letters do not differ significantly at $p=0.05$ within each of *Salmonella* and *Staphylococci* countMean value of control samples in total *Salmonella* count = $4.80 \times 10^2 \pm 3.2 \times 10^2$ Mean value of control samples in total *Staphylococcus aureus* count = $2.44 \times 10^2 \pm 67 \times 10^2$ Mean value of control samples in total Mold & yeast count = $2.93 \times 10^2 \pm 30 \times 10^2$

Hr = hour

Table 7: Incidence of Enterobacteriaceae, Staphylococcus aureus and Mould & yeasts isolated from examined samples of treated and untreated lamb carcasses (n=25)

Isolates	Control		Lactic A 1%		Lactic A 2%		Acetic A 1%		Acetic A 2%	
	N	%	N	%	N	%	N	%	N	%
<i>Edwardsiella tarda</i>	0	0	0	0	0	0	1	4	0	0
<i>Enterobacter aerogens</i>	3	12	1	4	0	0	0	0	1	4
<i>Enterob.agglomerance</i>	0	0	1	4	0	0	1	4	0	0
<i>E. coli</i>	5	20	5	20	1	4	2	8	0	0
<i>Khlebsiella oxytoca</i>	0	0	1	4	0	0	0	0	0	0
<i>Khleb.Pumonea. ozaene</i>	0	0	0	0	0	0	1	4	1	4
<i>Serratea rubidea</i>	1	4	0	0	0	0	1	4	0	0
<i>Salmonella enteritidis</i>	2	8	8	32	3	12	5	20	3	12
<i>Staphylococcus aureus</i>	5	20	23	92	14	68	22	88	16	64
Isolated Moulds										
<i>Aspergillus flavus</i>	0	0	0	0	1	4	1	4	1	4
<i>Aspergillus niger</i>	0	0	1	4	0	0	1	4	0	0
<i>Fusarium</i>	1	4	1	4	0	0	0	0	0	0
<i>Mucor spp.</i>	2	8	0	0	0	0	0	0	0	0
<i>Penicillium</i>	2	8	2	8	3	12	6	24	2	8

Table 8: Statistical analytical results of total proteolytic and lipolytic counts (cfu/g) of the examined lamb carcasses

Count	Treatment (means ± standard errors)									
	Control		Lactic acid 1%		Lactic acid 2%		Acetic acid 1%		Acetic acid 2%	
	% +ve	Mean ± SEM	% +ve	Mean ± SEM	% +ve	Mean ± SEM	% +ve	Mean ± SEM	% +ve	Mean ± SEM
Proteolytic (Reduction %)	92	38.19 ± 7.21 a	84	33.04 ± 6.29 ab (13.49)	84	22.05 ± 4.19 b (42.26)	88	29.41 ± 5.97 ab (22.99)	84	23.43 ± 5.80 b (38.65)
Lipolytic (Reduction %)	88	31.95 ± 1.85 a	88	21.45 ± 1.73 b (32.86)	88	12.88 ± 1.11 c (59.87)	88	21.77 ± 1.73 b (31.86)	88	12.50 ± 0.94 c (60.88)

Number of examined samples per treatment = 25.

Reduction % was the deviation of treatment means relative to the control mean.

Means in the same row followed by the same letter do not differ significantly P=0.05.

DISCUSSION

The present study aimed to assess the ability of organic acids to improve the quality of fresh lamb meat chilled at 7 °C for 24,48,72 hours, 7, 14 day and freezed at -18 °C for one month after treatment with 1,2% of both acetic and lactic acids. The gained results could be summarized as follow. pH value as illustrated in Table 1 revealed decline in pH values as 3.06 ± 0.05 and 3.06 ± 0.02 in treated samples with 2% acetic and lactic acids. after 24 hours of chilling, gradual increase was detected with time of chilling reached its maximum at 14 day of chilling and recording 6.08 ± 0.04 and 5.60 ± 0.08 in 1,2% lactic acid. Similar results were gained in acetic acid. These results agreed with those recorded by Kim (1988) who cited significant decrease in pH values of pork meat sprayed by acetic acid and stored at 4 °C for 12-15 day.

Total aerobic bacterial count: Table (2) showed a mean bacterial count value of the examined control lamb samples of $3.74 \times 10^3 \pm 3.88 \times 10^2$. After treatment with lactic acid 1% and acetic acid 1% it were $2.19 \times 10^3 \pm 2.25 \times 10^2$ and $2.06 \times 10^3 \pm 2.44 \times 10^2$. Using 2% lactic acid and acetic acid 2% the mean values declined to $8.8 \times 10^2 \pm 1 \times 10^2$ and $8.56 \times 10^2 \pm 1.08 \times 10^2$ c.fu /g. Successive decline in mean values of aerobic bacterial count with time of chilling after treatment reached in the 7th day to $6.38 \times 10^2 \pm 0.53 \times 10^2$ and $5.53 \times 10^2 \pm 0.51 \times 10^2$ and $5.96 \times 10^2 \pm 0.51 \times 10^2$ and $5.13 \times 10^2 \pm 0.5 \times 10^2$ cfu /g in lactic acid 1&2 % and acetic acid 1 and 2% respectively. While after 14day of chilling post treatment, slight increase was detected. The reduction percent of total aerobic bacterial count as illustrated in Table 3 revealed that, the best reduction percentages were 70.87, 64.09, 71.07 and 60.54% at 7th day chilling of lamb meat samples with lactic acid 1%,2% and acetic acid respectively this could be attributed to the destructive effect of these acids on different microbes, in addition to the longer acidic phase which started after bleeding of animal. Nearly similar results were reported by Anderson and Marshall (1989), Mendonca, *et al.* (1989) Gauthier and Jacquet (1991), Anderson *et al.* (1992), Zerby *et al.* (1999) and Mahmoud (2004). The freezing of treated samples for one month revealed the reduction percent varied from 62.37 to 62.53 57.96 and 57.38% respectively with using 1& 2% lactic acid and 1&2% acetic acid. That reduction percent could be attributed to the acid treatment

effect, in addition to the effect of freezing on different microbes. Nearly similar results were reported by Arjyapitipum *et al.* (1999).

Total psychrophilic bacterial count: Table 2 showed that, the mean value of total psychrophilic bacterial count (cfu/g) was $2.06 \times 10^3 \pm 2.85 \times 10^2$. Treatment with lactic acid 1% & 2% and acetic acid 1% & 2% revealed means as $1.53 \times 10^3 \pm 1.67 \times 10^2$, $1.2 \times 10^3 \pm 1.5 \times 10^2$ and $1.46 \times 10^3 \pm 1.82 \times 10^2$ and $1.02 \times 10^3 \pm 1.21 \times 10^2$ cfu/g respectively. The mean counts gradually decreased as after 7 days of chilling reached $7.72 \times 10^2 \pm 0.65 \times 10^2$, $6.8 \times 10^2 \pm 0.66 \times 10^2$ and $6.81 \times 10^2 \pm 0.72 \times 10^2$ and $6.32 \times 10^2 \pm 0.64 \times 10^2$ cfu/g respectively, more decline was recorded after 14 day of chilling of meat samples. This significant decrease of total psychrophilic count at $p = 0.05$ agreed with those reported by Anderson and Marshall (1989), Dorsa *et al.* (1998) and Lee *et al.* (1998).

Result in Table 4 denote that the reduction percent of psychrophilic count at the 7th day of chilling of treated lamb meat samples was the best reduction percent (49.54, 43.33, 53.36 and 38.04 %). Nearly similar results were reported by Anderson and Marshall (1989), Dickson, (1991) and Ariyapitipum *et al.* (1999).

Total coliform count: Table 4 Showed excessive decline with the time of storage at chilling after treatment with 1%, 2% lactic and acetic acids where in control the mean value was $1.53 \times 10^3 \pm 1.82 \times 10^2$ cfu/g. After treatment the means declined to $9.83 \times 10^2 \pm 1.31 \times 10^2$, $6.46 \times 10^2 \pm 0.99 \times 10^2$, $1.02 \times 10^2 \pm 1.41 \times 10^2$ and $4.6 \times 10^2 \pm 0.84 \times 10^2$ cfu/g respectively after 14 day of chilling post treated it reached to $4.63 \times 10^2 \pm 0.48 \times 10^2$, $3.93 \times 10^2 \pm 0.43 \times 10^2$ and $4.34 \times 10^2 \pm 0.43 \times 10^2$, $3.46 \times 10^2 \pm 0.43 \times 10^2$ cfu/g. Nearly similar results were recorded by Anderson and Marshall (1989), Castillo, *et al.* (1998), Dorsa *et al.* (1998) and Zerby *et al.* (1999)

The reduction percent as illustrated in Table 5 denoted that, at 7th day of chilling the reduction rate of the treated samples reached 73.65, 69.04, 77.84 and 68.59%. after application of lactic acid and acetic acid 1% & 2% respectively, while in freezing the reduction percent was 53.51, 45.51, 63.72 and 45.00%, respectively. Similar results were recorded by Anderson and Marshall (1989), Cutter and Siragusa (1994), Cabedo *et al.* (1996) and Ramirez, *et al.* (2001).

Total Enterobacteriaceae count: Table 4 revealed that, the mean value of control non treated sample was $2 \times 10^3 \pm 2.55 \times 10^3$ after treatment with 1%, 2% lactic acid acetic acid the reduction in mean values gradually increase till 7th day reached $3.69 \times 10^2 \pm 0.43 \times 10^2$, 3.25×10^2

$\pm 0.4 \times 10^2$, $3.58 \times 10^2 \pm 0.4 \times 10^2$ and $2.97 \times 10^2 \pm 0.38 \times 10^2$. The higher concentration 2% acetic acid showed significantly ($p < 0.05$) lower count compared to other acid treatment. Nearly similar results were recorded by Anderson *et al.* (1992) and Ariyapitipun *et al.* (1999). In the 7th day of chilling. The highest reduction percent 68.06, 65.24, 71.81, and 64.77% were recorded as in Table 5. Similar results reported by Mendonca, *et al.* (1989) and Anderson *et al.* (1992)

Table 7 revealed that, the isolated Enterobacteriaceae from the examined lamb carcasses at variable percentages were *Edwardsiella tarda*, *Enterobacter arerogenes*, *Enterobacter agglornerans*, *Escherichia coli*, *Klebsiella oxytocea*, *Klebsiella pneurnoniae sub.ozaene* and *Serratia rubide* in the control and treated samples as well as during. Some isolates were found in control samples as well as in treated one during chilling and freezing.

The presence of coliforms in the food pointed at the unsanitary condition of slaughter and processing plants as they are indicative of fecal pollution either from workers, lamb or all equipment kept in touch with them. Efforts should be directed towards thorough cleaning and sanitizing all equipment come in contact with lamb carcasses and workers, thorough cleaning of lamb carcasses and hygienic measures should be adopted during different stages.

Salmonella count: Table 6 shows the mean *Salmonella* count of 25 examined control lamb carcasses was $4.8 \times 10^2 \pm 3.2 \times 10^2$. After treatment with lactic acid 1% and 2%, acetic acid 1% & 2% the means of *Salmonella* count gradually decreased till reached $0.6 \times 10^2 \pm 0.00$, $0.2 \times 10^2 \pm 0.00$, $0.6 \times 10^2 \pm 0.2 \times 10^2$ and $0.4 \times 10^2 \pm 0.00$ respectively. After 72 hour of treatment reduction rate became 76.92, 83.33, 78.57 and 69.23%. Nearly similar results were reported by Anderson and Marshall, (1990), Anderson *et al.* (1992), Conner, *et al.* (1997) and Zerby *et al.* (1998)

Table 7 denote the identified *Salmonella enteritidis* isolated from the examined lamb carcasses incidentally appeared with its percentage as 2 (8%) in the control, while in lactic acid (1% and 2%) treated samples, the percentages were 8 (32%) and 3 (12%), respectively. In acetic acid (1% and 2%) treated samples it was shown as 5 (20%) and 3 (12%). *Salmonella* remains one of the most common causes of bacterial food poisoning and associated with the consumption of meat. These organisms colonize the

alimentary tract and excreted in the feces by infected animals thereby the human food chain. *Salmonella typhimurium* and *Salmonella enteritidis* are numerically the predominant serotypes affecting meat causing human infection through consumption of meat.

***Staphylococcus aureus* count:** Table 6 shows a mean value of $2.44 \pm 0.67 \times 10^2$ in examined control. After treating samples with 1% & 2% lactic acid and acetic acid means were gradually decreased with continuous chilling till 7th day reached to $0.8 \times 10^2 \pm 0.22 \times 10^2$, $0.45 \times 10^2 \pm 0.19 \times 10^2$, $0.6 \times 10^2 \pm 0.22 \times 10^2$ and $0.50 \times 10^2 \pm 0.3 \times 10^2$ cfu/g, respectively. That highest reduction forms 42.86, 60.86, 59.46, 33.33% as well as samples treated with 1% & 2% lactic acid and 1% & 2% acetic acid after freezing the treated samples the reduction percents as 7.14, 64.91, 55.41 and 60% respectively Table(3) Nearly similar results were reported by Mahmoud (2004)

Staphylococcus aureus could be isolated from the same examined samples as 23 (92%) lactic acid 1%, 14 (68%) lactic acid 2%, 22 (88%), acetic acid 1%, and 16 (64%) acetic acid 2%. *Staphylococci* are widespread in nature; they are members of the normal bacterial flora of the skin and mucous membranes. *Staphylococcus aureus* was frequently involved in case of mastitis and suppurative infections. Hence contamination of food with the organism mostly occurs due to sanitary neglected precautions during production or processing. When conditions are favorable for growth and multiplication of the organism in food, the enterotoxins are produced and consequently the food is likely to be dangerous. Enterotoxigenic *Staphylococcus aureus* has been implicated in several food poisoning outbreaks from consumption of lamb carcasses and its products (Beckers, 1982).

Total mould and yeast count: Table 6 revealed a mean of $2.93 \times 10^2 \pm 0.3 \times 10^2$ cfu/g, in control sample. When lactic acid 1% & 2%, acetic acid 1%, 2% were applied the mean count of mould and yeasts continued to decline with time of chilling reached to $0.77 \times 10^2 \pm 0.08 \times 10^2$, $0.58 \times 10^2 \pm 0.11 \times 10^2$, $0.72 \times 10^2 \pm 0.07 \times 10^2$ and $0.50 \times 10^2 \pm 0.06 \times 10^2$ respectively after 72 hour of chilling, the reduction percent of yeasts & mould as in Table 5 recorded as 56.74, 52.07, 52.94 and 39.02% respectively. Nearly similar results were recorded by Samelies *et al.* (2002). A slight rising of mean values was recorded after 7 and 14 day of chilling. This could be attributed to optimal pH of mould, yeast growth as recorded by Samelies *et al.* (2002) who mentioned that acids

containing washings were selective for growth of yeast. Slight decline recorded after freezing for one month. This was attributed to that mould and yeast grow at chilling and freezing as reported by Samelis *et al.* (2002). Table 7 also reveals that *Aspergillus flavus*, *Aspergillus niger*, *Fusarium*, *Mucor* and *Penicillium* could be isolated from the examined treated lamb samples at variable percentages. From the public health point of view, species of *Aspergillus* may induce pulmonary aspergillosis and allergy, and skin infection (Al-Doory, 1980 and Washington, 1981) for meat handlers. *Penicillium* spp. may induce pulmonary infection (Washington, 1981) *Mucor* and *Rhizopus* spp. are prevalent in food and may induce infection in lungs, gastrointestinal tract and skin (Al-Doory, 1980 and Washington, 1981).

Proteolytic and lipolytic bacterial counts: Both proteolytic and lipolytic bacterial counts on using lactic acid 1 and 2%, acetic acid 1&2% revealed reduction in their mean counts. The highest reduction percent was apparently with using lactic acid 2% as 42.26% and 59.87% for proteolytic and lipolytic bacteria, while that of acetic acid 2% was 38.65 and 60.88% respectively. These results agreed with that reported by Gill and Newton (1982) Anderson *et al.* (1988) Katoh *et al.* (1991) and Hassan (2001)

The isolated strains are of economic importance as they are food spoilage microorganisms besides they are of public health hazard, especially *E. coli* which causes acute infection to adult. It also causes infections of the urinary tract (Cruickshank, *et al.*, 1975).

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