EFFECT OF HOT WATER, ACETIC ACID, CHLORINE AND TRI-SODIUM PHOSPHATE ON MICROBIAL CONTAMINATION REDUCTION OF SOME MEAT BY-PRODUCTS.

By

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SUMMARY

Fifty meat by-product samples were collected from Cairo slaughterhouse (El.Bassatin), represented by lips, large intestine, liver, ox tail and tongue (10 from each). Samples were collected and bacteriologically examined for aerobic plate count, coliform count and Escherichia coli count immediately before and after immersion in different types of decontaminants such as hot water, acetic acid, chlorine and tri-sodium phosphate. The most effective treatment in reduction of aerobic plate count was acetic acid in case of lips and tongue,: tri-sodium phosphate in case of large intestine, while hot water in case of ox tail and chlorine in case of liver samples.

On the other hand, the most effective treatment for reducing the coliform count in lips, ox tail and tongue samples was the acetic acid, while mean tri-sodium phosphate was the most effective in large intestine and liver samples, but in case of Escherichia coli, hot water treatment was the highest reductant for contamination in lip and tongue samples, in large intestine and liver samples tri-sodium phosphate was the best decontaminant types, but in ox tail was acetic acid was the more effective type.

INTRODUCTION

The hygienic status of animal carcasses has long been a concern to the meat-processing industry. During beef slaughtering the carcass may become contaminated with different sources of bacteria including processing equipments, workers and surrounding environment as well as during evisceration from the internal viscera; however the predominant source of bacterial contamination is the animal itself. Dressing is the major hazard for carcass contamination with fecal bacteria. The characterization of

a process should then preferably involve the enumeration of an indicative organism for fecal contamination, such as coliform and *Escherichia coli* (*Mackey and Roberts, 1993*).

Numerous studies have evaluated technological processes for the decontamination of meat and meat by-products (*Brackett et al., 1994; Dorsa et al., 1996; Dorsa 1997; Phebus et al., 1997 and Delmore et al., 2000*). Decontamination of carcasses with organic acids and other chemical sanitizers has been extensively used (*Anderson and Marshall, 1990*).

Moreover, the use of cold and hot water as decontaminant treatments were recorded by *Hardin et al. (1995); Prasai et al. (1991) and Dorsa et al. (1996)*. Organic acids have been studied extensively for reducing bacterial populations on carcass surfaces and their effectiveness depends on concentration, temperature, exposure time, mode of application, type of meat tissue evaluated and sensitivity of specific bacterial populations (*Anderson and Marshall, 1990; Brackett et al., 1994; Cutter and Siragusa, 1994; Dickson and Siragusa, 1994 and Hardin et al., 1995*). Chlorine and tri-sodium phosphate were used by *Delmore et al. (2000)* as bacterial decontaminant treatments in meat and meat by-products.

So, the objective of this research was done to determine and compare the effectiveness of each of hot water, acetic acid, chlorine and tri-sodium phosphate on aerobic plate count, coliform count and *Escherichia coli* count on selective meat by- products collected from EL- Bassatin slaughterhouse.

MATERIALS and METHODS

Meat by-product samples:

Fifty beef by-product samples were collected from EL-Bassatin slaughterhouse including lips, large intestine, livers, ox tails and tongues (10 from each). Samples were selected due to their high probability of coliform and *Escherichia coli* contamination levels. The weight of each sample was about 200 gm. which was divided into two equal parts. The first part (control group) was subjected to bacterial count immediately after collection, the other part (treatment group) was subjected to different decontaminants, then bacterial counts were done.

Bacterial decontaminations (Delmore et al., 2000):

Each sample of the treatment group was subdivided into 4 parts; (each 25 gm). Each part was exposed to one type of different treatments used to reduce the microbial load of the meat by-product samples, included hot water (sterile distilled water) at 80°C ,acetic acid (2% vol/vol) with pH 2.8 and used at 50°C which prepared from glacial acetic acid, chlorine (sodium hypochlorite 0.005% wt/vol) with pH 6.5 and used at 50°C. and tri-sodium phosphate (12% wt/vol) with pH 12.5 and also used at 50°C which prepared from tri-sodium phosphate hydrate.

Treatment solutions (acetic acid, chlorine and tri-sodium phosphate) were prepared with sterile distilled water by mixing 2 liters of the appropriate solution in a sterile container and heated in a water bath (calibrated with thermometer) adjusted at 50°C When hot water and other treatment solutions reached the proper temperature, the various meat by-product samples were held with sterile forceps and immersed in the liquid for 10 seconds under aseptic conditions.

Immediately after decontaminant applications, samples were kept into sterile bags for 15 min. at room temperature before the bacteriological count. The holding time was applied to resemble the time that could exist in a commercial facilities between treatment applications and any method of meat by-product preservation.

Microbiological examinations:

Control and treatment group samples were analyzed for total aerobic counts, total coliform counts and *Escherichia coli* counts. Surface rinsing procedure was used to dislodge bacteria from the irregular surface of each meat by-product sample. A 225 ml. of peptone water 1% was added to each bag containing 25 gm. of sample to prepare ten fold serial dilutions.

Aerobic plate counts were done by spreading 0.1ml from each dilution on tryptic soya agar plates which were incubated for 48 hr. at 35°C (*Delmore et al., 2000*).

Total Coliform and Escherichia coli counts were done by surface-plating onto BCIG (5-Bromo-4-Chloro-3-indolyl-β-D-glucuronide) agar. Plates were incubated for (TCCs) at 35°C. for 24 hr. and for (ECCs) at 30°C for 4 hr and then at 44°C for 18 ± 2

hr. Identification was confirmed for *Escherichia coli* with a positive β glucuronidase reaction (*Frampton and Restaino, 1993*).

RESULTS

The obtained results were recorded in tables ,1,2,3 and figure 1.

DISCUSSION

It is evident from the results recorded in *table (1)* that the aerobic plate count reduction of lips samples treated with hot water, acetic acid, chlorine and tri-sodium phosphate was 1.24, 1.83, 0.40 and 1.20 log CFU/g, respectively, while in large intestine samples the reduction was 0.04, 0.60, 0.31 and 1.08 log CFU/g respectively. In liver samples, the reduction was 0.45, 0.82, 1.15 and 0.75 log CFU/g , while, in ox tail samples the reductions were 1.21, 1.15, 0.10 and 0.15 log CFU/g, but in tongue samples, APC was reduced by 1.40, 2.55, 0.15 and 0.55 log CFU/g for hot water acetic acid, chlorine and tri-sodium phosphate, respectively. These results were agree with those obtained by *Davey and Smith (1989); Prasai et al.(1991); Brackett et al.(1994); Hardin et al.(1995); Graves Delmore et al.(1998), Sofos and Smith (1998)* and *Delmore et al. (2000)* . On the other hand, from data obtained in *table (1)* after calculation found that the average reduction in Aerobic plate count for different types of meat by-products treated with acetic acid was 1.39 log CFU/g which agree with *Dorsa (1997*) who mentioned that the use of acetic acid 2% reduced the APC on beef samples by 1.3 - 2.0 log CFU/cm^2.

Moreover, the recorded results cleared that in lips and tongue samples the most effective treatment was acetic acid ,while in large intestine samples was tri-sodium phosphate, but in ox tail samples was hot water and finally in liver was chlorine. The greater reduction in bacterial populations were recorded when the temperature of the tri-sodium phosphate solution was increased to *Dickson et al., 1994*).

The results obtained in table (2) revealed that coliform counts were reduced in lips samples immersed in hot water acetic acid, chlorine and tri-sodium phosphate by 1.00, 2.05, 0.18 and 1.80 log CFU/g, respectively, while In large intestine samples the total coliform counts were reduced by 0.67, 1.35, 0.03 and 1.90 log CFU/g, respectively, while mean in liver samples were reduced by

0.76, 1.19, 0.60 and 1.38 log CFU/g respectively. However, in ox tail samples total coliform counts reduction was 1.57, 2.37, 0.52 and 2.07 log CFU/g, respectively. While in tongue samples the reductions were 1.10, 2.35, 0.30, 1.66 log CFU/g, in hot water, acetic acid, chlorine and tri-sodium phosphate ,respectively. The results recorded in this study were nearly similar to those obtained by *Delmore et al. (2000)*. It is interesting to note that the most effective treatments were the acetic acid in lips, ox tail and tongue samples, and tri-sodium phosphate in large intestine and liver samples.

It is evident from the results recorded in table (3) that the *Escherichia coli* count was reduced in lips samples immersed in hot water, acetic acid, chlorine and tri-sodium phosphate by 1.58, 1.28, 0.23 and 1.47 log CFU/g, respectively. In Large intestine the *Escherichia coli* counts were reduced by 0.59, 1.57, 0.02 and 2.00 log CFU/g, respectively. Concerning liver samples the values were reduced by 0.58, 1.49, 0.80 and 1.50 log CFU/g, respectively. While mean, in ox tail samples it was reduced by 1.39, 2.22, 0.11 and 2.14 log CFU/g, respectively, and in tongue *Escherichia coli* counts reduction were 0.60, 0.59, 0.10 and 0.41 log CFU/g, in hot water, acetic acid, chlorine and tri-sodium phosphate, respectively. Obtained results resembled those recorded by *Delmore et al. (2000)*. Data in table (3) showed that the most effective treatment was the hot water in lips and tongue samples while in large intestine and liver samples was tri-sodium phosphate and finally in ox tail samples was acetic acid.

Concerning the stated results illustrated in figure (1) it is clear that the acetic acid treatment was the most effective treatment reduced aerobic plate count, total coliform count and *Escherichia coli* count in each of lips, ox tail and tongue samples, while the tri-sodium phosphate was the best decontaminant for reduction of aerobic plate count, total coliform count and *Escherichia coli* count in each of large intestine and liver samples. *Bacon et al. (2000)* mentioned that total bacterial count, Coliform count and *Escherichia coli* counts on carcass surfaces after hide removal were ranged from 6.1 to 9.1; 3.0 to 6.0 and 2.6 to 5.3 log CFU/100 cm ^{2}, respectively. After decontamination by washing carcass with organic acids and hot water, the log mean of TPC, TCC and ECC on carcass surfaces were reduced to 3.8 - 7.1, 1.5 - 3.7 and 1.0 - 3.0 CFU/100 cm^{2} respectively.

In general, the highest efficacy of hot water on reduction of bacterial population was in lips and ox tail samples and less efficacy in other samples. *Castillo et al. (1998)* mentioned that, the efficacy of hot water treatments was affected by the carcass surface region, but not by delaying of the treatment time. Also, the highest efficacy of acetic acid treatment was in lips, ox tail and tongue samples, but chlorine was in liver samples; and tri-sodium phosphate was in lips, large intestine and ox tail samples.

Finally, it may be concluded that, the highest efficacy of different treatments for selected meat by-products samples were acetic acid, tri-sodium phosphate, hot water and finally chlorine, respectively. The lack of efficacy of chlorine for reduction of microbial populations may be attributed to the high levels of organic matter on meat by-products samples which inactivate the chlorine (*Cutter and Siragusa, 1994*).

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Table (1)The log mean values (CFU/g) of Aerobic plate counts of selected meat by-products treated with some decontaminants (n=10).

Treatments	Meat by-products														
	Lips			Large intestine			Liver			Ox tail			Tongue		
	B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%
Hot water	5.89	1.24	21	4.96	0.04	1	3.85	0.45	12	5.56	1.21	22	5.90	1.40	24
Acetic acid		1.83	31		0.60	12		0.82	21		1.15	21		2.55	43
Chlorine		0.40	7		0.31	6		1.15	30		0.10	2		0.15	3
Tri-sodium phosphate		1.20	20		1.08	22		0.75	19		0.15	3		0.55	9

B. = Counts before treatment **R. = Counts reduction after treatment**

Table (2): The log mean values CFU/g of Coliform counts of selected meat by-products treated with some decontaminants (n=10).

Treatments		Meat by-products											
		Lips			Large intestine			Liver			Ox tail			Tongue		
		B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%
Hot water		3.55	1.00	28	3.07	0.67	22	3.36	0.76	23	4.39	1.57	36	2.70	1.10	41
Acetic acid			2.05	58		1.35	44		1.19	35		2.37	54		2.35	87
Chlorine			0.18	5		0.03	1		0.60	18		0.52	12		0.30	11
Tri-sodium phosphate			1.80	51		1.90	62		1.38	41		2.07	47		1.66	61

Table (3): The log mean values of Escherichia coli counts CFU/g) of selected meat by-products treated with some decontaminants (n=10).

Treatments		Meat by-products											
		Lips			Large intestine			Liver			Ox tail			Tongue		
		B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%	B.	R.	%
Hot water		3.08	1.58	51	2.59	0.59	23	2.88	0.58	20	3.22	1.39	43	1.34	0.60	45
Acetic acid			1.28	42		1.57	61		1.49	52		2.22	69		0.59	44
Chlorine			0.23	7		0.02	1		0.80	28		0.11	3		0.10	7
Tri-sodium phosphate			1.47	48		2.00	77		1.50	52		2.14	66		0.41	31

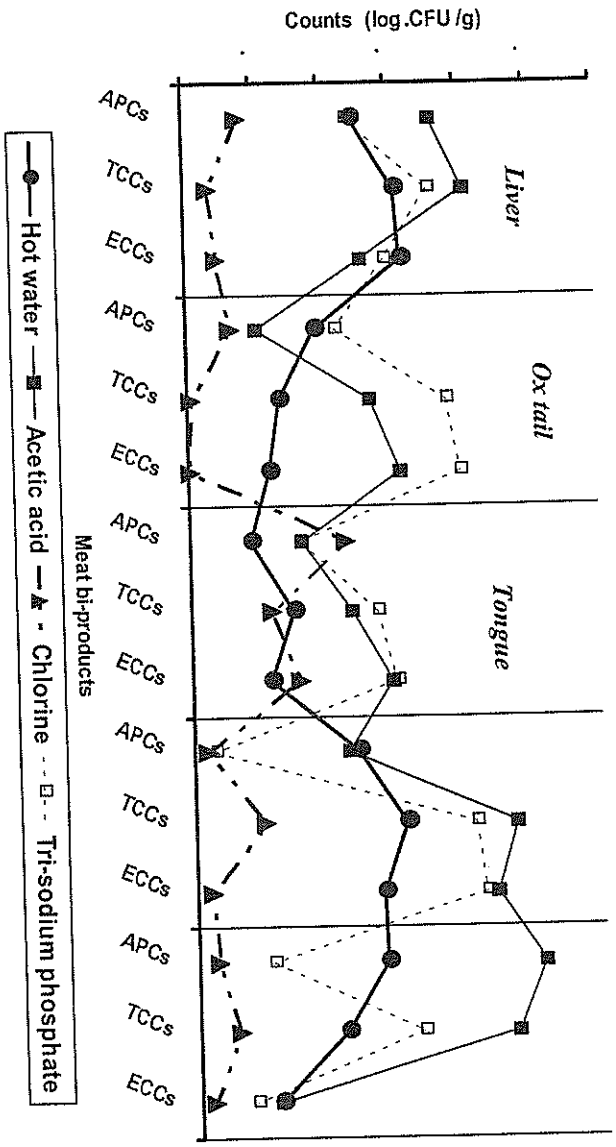

Figure ( ): The reduction of Aerobic plate, Coliform and Escherichia coli counts in selected meat by-products treated with some

الملخص العربي

تأثير الماء الساخن ، حمض الخليك ، الكلور و ثلاثي فوسفات الصوديوم على خفض التلوث الميكروبي لبعض مخلفات اللحوم

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معهد بحوث صحة الحيوان – *معمل الجيزة– **معمل طنطا

تم جمع خمسون عينة من مخلفات اللحوم من مجزر البساتين بالقاهرة ممثلة في الشفتين، الأمعاء الغليظة، الكبد، الذيل واللسان (10 من كل نوع). تم فحص العينات بكتريولوجيا للعدد الكلي للبكتيريا الهوائية، الكوليفورم والاشيريشيا كولاي بعد جمع العينات مباشرة ثم بعد غمرها في أنواع مختلفة من مزيلات التلوث البكتيري مثل الماء الساخن، حمض الخليك، الكلور، ثلاثي فوسفات الصوديوم.

وقد اثبتت النتائج ان حمض الخليك الأقوى تأثيرا فى خفض العد الكلي للبكتيريا الهوائية في عينات الشفتين واللسان ، ثلاثي فوسفات الصوديوم في عينات الأمعاء الغليظة،بينما كان الماء الساخن في عينات الذيل والكلور في عينات الكبد. ومن ناحية أخرى كان حمض الخليك أقوى تأثيرا فى خفض العد الكلي للكوليفورم في عينات الشفتين، الذيل واللسان, بينما كان ثلاثي فوسفات الصوديوم هو الأكثر تأثيرا في عينات الأمعاء الغليظة والكبد ، أما بالنسبة لميكروب الإشيريشيا كولاي كان الماء الساخن هو الأكثر خفضا للعدد الكلي للميكروب في عينات الشفتين واللسان، ثلاثي فوسفات الصوديوم في عينات الأمعاء الغليظة والكبد بينما في عينات الذيل كان الأكثر تأثيرا هو حمض الخليك.

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