



UNIVERSITI PUTRA MALAYSIA

**DECONTAMINATION OF CHICKEN BREASTS USING ORGANIC
ACIDS AND LAURICIDIN**

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FSTM 2005 2

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AND LAURICIDIN**

By

DANIEL MENSAH ANANG

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,
in Fulfilment of the Requirement for the Degree of Doctor of Philosophy**

December 2005



*Dedicated to
my beloved wife (Nancy Anang)
and
daughter (Aniyanni Yehowahi Naa Ode Anang)*



Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Doctor of Philosophy

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Chairman: Professor Gulam Rusul Rahmat Ali, PhD

Faculty: Food Science and Technology

Lauricidin, lactic and oxalic acids were evaluated for their effectiveness in reducing and inhibiting the growth of predominant spoilage and pathogenic microorganisms. Chicken breasts (150 – 200 g each) of freshly slaughtered chickens were purchased from a local wet market and analysed within 2 hr. Chicken breasts were dipped in 0, 0.5, 1.0, 1.5 and 2.0% solutions of lauricidin (w/v, containing 1% lactic acid and 1% ethanol), lactic acid (v/v) or oxalic acid (w/v) for 10, 20 and 30 min, then individually packed in oxygen-permeable polyethylene bags, and stored at 4°C. Aerobic plate counts (APC), populations of *Pseudomonas* spp. and *Enterobacteriaceae* on chicken breasts were determined before, after treatment and after storage for 1, 3, 7, 10, and 14 days at 4°C. Surviving aerobic organisms were isolated and identified from chicken breasts treated with lauricidin, lactic and oxalic acids. Dipping chicken breasts in solutions of lauricidin, lactic and oxalic acids caused significant ($P \leq 0.05$) reduction in APC and also retarded microbial growth throughout the 14 d storage period. APC on chicken samples treated with 0.5 to 2.0% lauricidin, lactic acid and oxalic acid solutions were significantly ($P \leq 0.05$) reduced by 0.92 - 1.2, 0.53-2.36 and 1.38-2.76 log CFU/g, respectively. Initial



Pseudomonas counts on samples treated with 0.5 to 2.0% lauricidin and lactic acid were in the range of 0.79 – 1.77 and 0.39 – 1.82 log CFU/g, respectively, which were significantly ($P \leq 0.05$) lower compared to fresh samples, and growth of *Pseudomonas* spp. was limited throughout the storage period. In chicken breasts treated with 0.5 to 2.0% lauricidin and lactic acid, *Enterobacteriaceae* counts decreased by 0.14–1.14 and 0.59-2.18 log CFU/g, respectively. Less than log 2 CFU/g *Enterobacteriaceae* and *Pseudomonas* counts were observed on samples treated with 1.0 – 2.0% oxalic acid for 10 to 30 min. *Enterobacter cloacae*, *Pseudomonas lundensis* and *Kocuria rhizophila* were the predominant aerobic organisms isolated from chicken breasts treated with 0.5 to 2.0% lauricidin, lactic acid and oxalic acid, respectively.

Lauricidin, lactic and oxalic acids were also evaluated for their effects on growth and survival of *Listeria monocytogenes* (L55), *Salmonella* Enteritidis (S552) and *Escherichia coli* O157:H7 (E19) inoculated onto raw chicken breasts. In chicken breasts treated with 0.5 to 2.0% lauricidin, initial counts of *L. monocytogenes*, *S. Enteritidis* and *E. coli* O157:H7 were significantly ($P \leq 0.05$) reduced by 2.90, 1.31 and 2.27 log CFU/g, respectively. *L. monocytogenes*, *S. Enteritidis* and *E. coli* O157:H7 counts on samples treated with lactic acid were significantly ($P \leq 0.05$) reduced by 1.97, 1.71 and 2.59 log CFU/g, respectively. Initial counts of *L. monocytogenes*, *S. Enteritidis* and *E. coli* O157:H7 in chicken samples treated with oxalic acid were significantly ($P \leq 0.05$) reduced by 2.87, 2.02 and 4.12 log CFU/g, respectively. Dipping of chicken breasts in higher concentrations of solutions of lauricidin, lactic and oxalic acids and longer dipping time gave additional benefit.

The colour and pH of chicken breasts dipped in solutions of lauricidin, lactic acid and oxalic acid were also evaluated. Sensory attributes of chicken breasts dipped in oxalic acid were determined. Oxalic acid residues in chicken breasts treated with oxalic acid were also determined. Dipping of chicken breasts in 0.5 to 2.0% lauricidin, lactic acid and oxalic acid caused significant ($P \leq 0.05$) decreased in pH, however, the decrease in pH was more pronounced in samples dipped in oxalic acid. Dipping chicken breasts in 0.5 to 2.0% lauricidin, lactic acid and oxalic acids caused slight darkening, as reflected by the increase in Hunter *L* values. Lauricidin caused a slight decrease in Hunter *a* value (decreased redness), and an increase in Hunter *b* value (increase in yellowness). Lactic acid also caused an increase in Hunter *a* values (increased redness) and Hunter *b* values (increased yellowness). Oxalic acid gave a more bleached chicken breasts compared to lauricidin and lactic acid. Higher concentrations of oxalic acid and longer dipping time caused more bleaching of the chicken breasts compared to lower concentrations and shorter dipping time. The maximum residue of oxalic acid in unwashed chicken breasts was 36 mg/100g (in chicken samples dipped in 2% oxalic acid for 30 min). Oxalic acid residue significantly ($P \leq 0.05$) decreased by more than 50% when the chicken breasts were washed and subsequently cooked. The maximum residue of oxalic acid in roasted chicken breast was 2 mg/100g. Results showed that lauricidin, lactic and oxalic acids have the potential to be used as a sanitizer in chicken carcasses during processing. Sensory evaluation of chicken breasts treated with oxalic acid demonstrated that, even though instrumental measurements may indicate deterioration in appearance, cooked chicken breasts were acceptable to consumers.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

**DEKONTAMINASI DADA AYAM MENGGUNAKAN ASID ORGANIK
DAN LAURICIDIN**

oleh

DANIEL MENSAH ANANG

Disember 2005

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Penilaian terhadap keberkesanan lauricidin, asid laktik dan asid oksalik untuk mengurangkan dan merencatkan pertumbuhan mikroorganisma perosak pradominan dan patogenik telah dilakukan. Bahagian dada ayam (150 – 200 g setiap satu) daripada ayam yang baru disembelih dibeli daripada pasar tempatan dan dianalisis dalam masa 2 jam. Bahagian dada ayam dicelup ke dalam 0, 0.5, 1.0, 1.5 and 2.0% larutan lauricidin (w/v, mengandungi 1% asid laktik dan 1% etanol), asid laktik (v/v) atau asid oksalik (w/v) selama 10, 20, and 30 min, dan kemudian dibungkus secara berasingan di dalam beg polietilena yang telus oksigen dan disimpan pada suhu 4°C. Aerobic Plate Counts (APC), populasi *Pseudomonas* spp. dan *Enterobacteriaceae* pada dada ayam ditentukan sebelum dan selepas rawatan serta selepas disimpan selama 1, 3, 7, 10, dan 14 hari pada 4°C. Organisma yang dapat terus bertahan dan hidup pada dada ayam setelah dirawat dengan asid laktik, asid oksalik dan lauricidin telah diasingkan dan dikenalpasti. Dada ayam yang dicelup dalam lauricidin, asid laktik dan asid oksalik menyebabkan pengurangan bererti ($P \leq 0.05$) APC dan merencat pertumbuhan bakteria sepanjang 14 hari tempoh penyimpanan. APC pada sampel ayam yang telah dicelup dalam 0.5 hingga 2.0% larutan lauricidin, asid laktik



dan asid oksalik masing-masing berkurangan secara bererti ($P \leq 0.05$) sebanyak 0.92 - 1.2, 0.53-2.36 dan 1.38-2.76 log CFU/g. Rawatan dengan lauricidin telah dapat mengurangkan kiraan *Pseudomonas* sebanyak 0.79 – 1.77 log CFU/g manakala rawatan dengan asid laktik mengurangkan kiraan *Pseudomonas* sebanyak 0.39 – 1.82 log CFU/g, di mana pertumbuhan ini sangat terhad sepanjang tempoh simpanan dan berkurangan secara bererti ($P \leq 0.05$) jika dibandingkan dengan pertumbuhan pada sampel ayam segar. Kiraan *Enterobacteriaceae* pada sampel ayam yang dicelup dalam 0.5 hingga 2.0% larutan lauricidin berkurangan sebanyak 0.14–1.14 log CFU/g manakala kiraan pada sampel ayam yang dicelup dalam 0.5% hingga 2.0% larutan asid laktik berkurangan sebanyak 0.59-2.18 log CFU/g. Kiraan *Enterobacteriaceae* dan *Pseudomonas* telah berkurangan sebanyak kurang daripada log 2 CFU/g apabila sampel ayam dicelup dalam larutan 1.0 – 2.0% asid oksalik selama 10 hingga 30 min. Bakteria yang mendominasi bahagian dada ayam yang telah dicelup dalam 0.5 hingga 2.0% lauricidin ialah *Enterobacter cloacae*. *Pseudomonas lundensi* pula mendominasi bahagian dada ayam yang telah dicelup dalam 0.5 hingga 2.0% asid laktik manakala *Kocuria rhizophila* mendominasi bahagian dada ayam yang telah dicelup dalam 0.5 hingga 2.0% asid oksalik.

Keberkesanan lauricidin, asid laktik dan asid oksalik untuk merencat pertumbuhan *Listeria monocytogenes* (L55), *Salmonella* Enteritidis (S552) dan *Escherichia coli* O157:H7 (E19) yang telah diinokulkan ke atas dada ayam diuji. Kiraan awal *L. monocytogenes* berkurangan secara bererti ($P \leq 0.05$) sebanyak 2.90 log CFU/g, kiraan awal *S. Enteritidis* berkurangan secara bererti ($P \leq 0.05$) sebanyak 1.31 log CFU/g dan kiraan awal *E. coli* O157:H7 berkurangan secara bererti ($P \leq 0.05$) sebanyak 2.27 log CFU/g apabila sampel ayam dirawat dengan 0.5 hingga 2.0%

larutan lauricidin. Asid laktik mengurangkan kiraan awal *L. monocytogenes*, *S. Enteritidis* dan *E. coli* O157:H7 secara bererti ($P \leq 0.05$) sebanyak 1.97, 1.71 dan 2.59 log CFU/g masing-masing. Kiraan awal *L. monocytogenes*, *S. Enteritidis* dan *E. coli* O157:H7 pada sampel ayam yang dicelup dalam larutan asid oksalik berkurangan secara bererti ($P \leq 0.05$) sebanyak 2.87, 2.02 dan 4.12 log CFU/g masing-masing. Dada ayam yang dicelup di dalam larutan lauricidin, asid laktik dan asid oksalik yang berkepekatan tinggi dan tempoh mencelup yang lebih lama dapat memberikan kesan yang lebih positif.

Warna dan pH dada ayam yang telah dicelup di dalam larutan lauricidin, asid laktik dan asid oksalik juga dinilai. Penilaian deria dan sisa asid oksalik pada dada ayam yang telah dicelup dengannya turut ditentukan. Mencelup dada ayam di dalam 0.5 hingga 2.0% larutan lauricidin, asid laktik dan asid oksalik memberi kesan yang signifikan ($P \leq 0.05$) pada pengurangan pH tetapi pengurangan pH lebih ketara pada dada ayam yang dicelup dengan asid oksalik. Rawatan menggunakan 0.5 hingga 2.0% larutan lauricidin, asid laktik dan asid oksalik menyebabkan warna menjadi sedikit gelap seperti yang ditunjukkan pada pertambahan nilai Hunter L. Larutan lauricidin menyebabkan terdapat sedikit pengurangan pada nilai Hunter a (pengurangan warna merah), dan peningkatan nilai Hunter b (peningkatan warna kuning). Larutan asid laktik menyebabkan peningkatan nilai Hunter a (peningkatan warna merah) dan nilai Hunter b (peningkatan warna kuning). Larutan asid oksalik menghasilkan produk yang lebih putih jika dibandingkan dengan larutan lauricidin dan larutan asid laktik. Kepekatan asid oksalik yang tinggi dan masa yang lebih lama semasa mencelup menghasilkan produk yang lebih putih jika dibandingkan dengan kepekatan asid oksalik yang rendah dan masa yang singkat semasa mencelup. Sisa

maksimum asid oksalik pada bahagian dada ayam yang belum dibasuh ialah 36 mg/100 g (sample ayam yang dicelup dalam 2% asid oksalik selama 30 min). Sisa asid oksalik berkurangan bereti 50% ($P \leq 0.05$) atau lebih apabila sampel dibasuh dan seterusnya dimasak. Sisa maksimum asid oksalik di dalam ayam yang telah dipanggang ialah 2mg/100g. Keputusan menunjukkan bahawa lauricidin, asid laktik, dan asid oksalik mempunyai potensi untuk digunakan sebagai bahan sanitasi untuk mengurangkan populasi mikroorganisma perosak dan patogenik yang wujud secara semulajadi semasa pemprosesan ayam mentah. Penilaian deria menunjukkan bahawa pengguna masih boleh menerima ayam yang telah dicelup di dalam asid oksalik walaupun sukatan menggunakan instrumentasi mungkin telah menunjukkan kemerosoton nilai dari segi rupa luaran.

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Daniel Mensah Anang

November 2005



I certify that an Examination Committee met on 30th December 2005 to conduct the final examination of Daniel Mensah Anang on his Doctor of Philosophy thesis entitled “Decontamination of Chicken Breasts Using Organic Acids and Lauricidin” in accordance with University Pertanian Malaysia (Higher Degree) Act 1980 and University Pertanian Malaysia (Higher Degree) Regulations 1981. The Committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follows:

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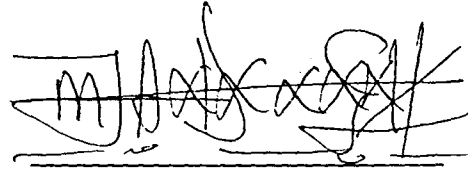
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DECLARATION

I hereby declare that the thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.



DANIEL MENSAH ANANG

Date: 17th January 2006

TABLE OF CONTENTS

	Page
DEDICATION	ii
ABSTRACT	iii
ABSTRAK	vi
ACKNOWLEDGEMENTS	x
APPROVAL	xii
DECLARATION	xiv
LIST OF TABLES	xviii
LIST OF FIGURES	xxii
 CHAPTER	
 1 INTRODUCTION	 1
 2 LITERATURE REVIEW	 7
2.1 Introduction	7
2.2 Microbiology of Poultry Slaughtering	9
2.2.1 Preslaughter Handling and Transportation	10
2.2.2 Scalding	11
2.2.3 Plucking (Defeathering)	12
2.2.4 Evisceration	14
2.2.5 Chilling	14
2.3 Spoilage of Poultry	15
2.3.1 Spoilage Organisms Associated with Poultry	15
2.3.2 Biochemistry and Characteristics of spoilage	17
2.4 Microbial Safety of Poultry	20
2.5 Control of Microorganisms in Poultry	33
2.5.1 Decontamination of Poultry	34
2.5.1.1 Non-chemical Decontamination	34
2.5.1.2 Chemical Decontamination	36
2.5.2 General Hygiene and Sanitation	47
2.5.3 Application of Quality Assurance Systems (GMP, GHP and HACCP) in Poultry Slaughtering	50
 3 INHIBITORY EFFECT OF LAURICIDIN, LACTIC AND OXALIC ACIDS ON SPOILAGE ORGANISMS OF RAW, CHILLED CHICKEN	 53
3.1 Introduction	53
3.2 Materials and Methods	55
3.2.1 Chemical and bacteriological media	55
3.2.2 Treatment of chicken breast with organic acids and lauricidin	55
3.2.3 Microbiological sampling and analysis	56
3.2.4 Isolation and identification of surviving bacteria	57
3.2.5 Statistical Analysis	57



3.3	Results	58
3.3.1	Microbial growth on chicken breast treated with Lauricidin	58
3.3.2	Microbial growth on chicken breast treated with lactic acid	67
3.3.3	Microbial growth on chicken breast treated with oxalic acid	74
3.4	Discussion	81
4	INHIBITORY EFFECTS OF LAURICIDIN, LACTIC AND OXALIC ACIDS ON <i>Listeria monocytogenes</i>, <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 IN RAW, CHILLED CHICKEN	87
4.1	Introduction	87
4.2	Materials and methods	88
4.2.1	Chemicals and bacteriological media	88
4.2.2	Preparation of bacterial inoculum	89
4.2.3	Chicken inoculation and treatment application	89
4.2.4	Microbiological sampling and analysis	90
4.2.5	Statistical analysis	91
4.3	Results	92
4.3.1	Inhibitory effects of lauricidin on <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 in raw, chilled chicken	92
4.3.2	Inhibitory effects of lactic acids on <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 in raw, chilled chicken	99
4.3.3	Inhibitory effects of oxalic acid on <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 in raw, chilled chicken	106
4.4	Discussion	114
5	EFFECTS OF LAURICIDIN, LACTIC AND OXALIC ACIDS ON pH, COLOUR AND SENSORY CHARACTERISTICS AND DETERMINATION OF OXALIC ACID RESIDUE IN CHICKEN BREAST	121
5.1	Introduction	121
5.2	Materials and Methods	122
5.2.1	pH Determination	122
5.2.2	Determination of colour	123
5.2.3	Determination of oxalic acid residue	123
5.2.4	Sensory characteristics of chicken breast treated with oxalic acid	124
5.2.5	Statistical Analysis	126
5.3	Results	126

5.3.1	Sensory characteristics of chicken breast treated with oxalic acid	126
5.3.2	pH Changes of chicken breast treated with lauricidin, lactic and oxalic acids	130
5.3.3	Colour changes in chicken breast treated with lauricidin, lactic and oxalic acids	135
5.3.4	Oxalic Acid Residue in Chicken Breast	148
5.4	Discussions	149
6	CONCLUSION AND RECOMMENDATIONS	152
	REFERENCES	155
	APPENDICES	175
	BIODATA OF THE AUTHOR	189



LIST OF TABLES

Table		Page
1	Numbers of <i>Salmonella</i> and <i>Campylobacter</i> (log MPN/carcass) on chicken carcasses scalded at three different temperatures.	12
2	<i>Pseudomonas</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20, and 30 min and stored at 4°C for 14 days	63
3	<i>Enterobacteriaceae</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20, and 30 min and stored at 4°C for 14 days	65
4	Identification of microorganisms isolated from chicken breast treated with lauricidin and stored at 4°C for 14 days	66
5	<i>Pseudomonas</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20, and 30 min and stored at 4°C for 14 days	70
6	<i>Enterobacteriaceae</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20, and 30 min and stored at 4°C for 14 days	72
7	Identification of microorganisms isolated from chicken breast treated with lactic acid stored at 4°C for 14 days	73
8	<i>Pseudomonas</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20, and 30 min and stored at 4°C for 14 days	77
9	<i>Enterobacteriaceae</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20, and 30 min and stored at 4°C for 14 days	79
10	Identification of microorganisms isolated from chicken breast treated oxalic acid stored at 4°C for 14 days	80
11	Log-reduction of <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 inoculated onto chicken breast upon dipping in different concentrations of lauricidin.	93



12	<i>Listeria monocytogenes</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20, and 30 min and stored at 4°C for 14 days	94
13	<i>Salmonella</i> Enteritidis Counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20, and 30 min and stored at 4°C for 14 days	96
14	<i>E. coli</i> O157:H7 counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20, and 30 min and stored at 4°C for 14 days	98
15	Log-reduction of <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 inoculated onto chicken breast upon dipping in different concentrations of lactic acid.	99
16	<i>Listeria monocytogenes</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20, and 30 min and stored at 4°C for 14 days	102
17	<i>Salmonella</i> Enteritidis counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20, and 30 min and stored at 4°C for 14 days	104
18	<i>E. coli</i> O157:H7 counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 min and stored at 4°C for 14 days	105
19	Log-reduction of <i>Listeria monocytogenes</i> , <i>Salmonella</i> Enteritidis and <i>Escherichia coli</i> O157:H7 inoculated onto chicken breast upon dipping in different concentrations of oxalic acid.	106
20	<i>Listeria monocytogenes</i> counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	109
21	<i>Salmonella</i> Enteritidis counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	111
22	<i>E. coli</i> O157:H7 counts on chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	113
23	Colour, flavour and odour scores of chicken breast treated with 0, 0.5 and 1.5% oxalic acid for 10 min and stored at 4°C for 14 days	128



24	Juiciness, texture and overall acceptability scores of chicken breast treated with 0, 0.5 and 1.5% oxalic acid for 10 min and stored at 4°C for 14 days	129
25	pH of lauricidin, lactic and oxalic acid solutions	130
26	<i>pH</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 minutes and stored at 4°C for 14 days	132
27	<i>pH</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	133
28	<i>pH</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	134
29	Hunter <i>L</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 minutes and stored at 4°C for 14 days	136
30	Hunter <i>a</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 minutes and stored at 4°C for 14 days	137
31	Hunter <i>b</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 minutes and stored at 4°C for 14 days	139
32	Hunter <i>L</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	140
33	Hunter <i>a</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	142
34	Hunter <i>b</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	143
35	Hunter <i>L</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	144
36	Hunter <i>a</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	146
37	Hunter <i>b</i> values of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 minutes and stored at 4°C for 14 days	147
38	Oxalic acid residue (mg/100 g) in chicken breast treated with 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min.	148



39	MicroLog results of bacteria identification of lauricidin treated samples	175
40	MicroLog results of bacteria identification of lactic acid treated samples	176
41	MicroLog results of bacteria identification of oxalic acid treated samples	177
42	Sample of 9-point scale questionnaire used in Sensory Evaluation	178
43	Total Count of <i>Listeria monocytogenes</i> inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 min and stored at 4°C for 14 days	180
44	Total count of <i>Salmonella</i> Enteritidis inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 min and stored at 4°C for 14 days	181
45	Total Count of <i>E. coli</i> O157:H7 inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10, 20 and 30 min and stored at 4°C for 14 days	182
46	Total Count of <i>Listeria monocytogenes</i> inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 min and stored at 4°C for 14 days	183
47	Total Count of <i>Salmonella</i> Enteritidis inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 min and stored at 4°C for 14 days	184
48	Total Count of <i>E. coli</i> O157:H7 inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10, 20 and 30 min and stored at 4°C for 14 days	185
49	Total Count of <i>Listeria monocytogenes</i> inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	186
50	Total Count of <i>Salmonella</i> Enteritidis inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	187
51	Total Count of <i>E. coli</i> O157:H7 inoculated chicken breasts treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10, 20 and 30 min and stored at 4°C for 14 days	188



LIST OF FIGURES

Figure		Page
1	Conventional processing of chicken and turkeys	8
2	Microbial inhibitions by weak organic acids	44
3	Changes in Aerobic Plate Count of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 10 min and stored at 4°C for 14 days.	59
4	Changes in Aerobic Plate Count of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 20 min and stored at 4°C for 14 days.	60
5	Changes in Aerobic Plate Count of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lauricidin for 30 min and stored at 4°C for 14 days.	61
6	Changes in Aerobic Plate Counts of chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 10 min and stored at 4°C for 14 days.	67
7	Changes in Aerobic Plate Counts on chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 20 min and stored at 4°C for 14 days.	68
8	Changes in Aerobic Plate Counts on chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% lactic acid for 30 min and stored at 4°C for 14 days.	69
9	Changes in Aerobic Plate Counts on chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 10 min and stored at 4°C for 14 days.	74
10	Changes in Aerobic Plate Counts on chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 20 min and stored at 4°C for 14 days.	75
11	Changes in Aerobic Plate Count on chicken breast treated with 0, 0.5, 1.0, 1.5 and 2.0% oxalic acid for 30 min and stored at 4°C for 14 days.	76
12	Standard plot for oxalic acid concentrations in 2.0 HCl	179



CHAPTER 1

INTRODUCTION

Chicken forms about two thirds of the total world poultry meat production (Anon, 1996). Poultry meat generally, is perceived as cheap, wholesome, and nutritious, being high in protein and low in fat. There are no religious constraints on consumption, unlike pork and beef (Mead, 2000). These characteristics and other factors has made poultry meat by far the most popular food product worldwide (Mulder, 1999). Poultry meat production has increased rapidly worldwide over the years and this increase is associated with equivalent increase in intensive animal production with both increase in the number of farms and flock size (Bolder, 1998). It was reported by the United States Food and Agricultural Policy Research Institute (FAPRI) (2005) that major disease outbreaks in both beef and pork industries shifted consumption in many countries toward poultry. In the long run, however, lower cost and health considerations are driving the increase in world poultry consumption. The per capita consumption of poultry for 2003 were estimated as 50.1 kg in the USA, 35.9 kg in New Zealand, 34.2 kg in Australia, 22.7 kg in Western Europe, 37.1 kg in Canada and 32.5 kg in Malaysia (year 2002) (MAFnet, 2003; USDA, 2002). FAPRI (2005) projects that over the next 10 years, per capita consumption, production, and broiler net trade will grow at annual rates of 2.1, 3.2 and 3.7 percent, respectively. It was estimated that global trade in broiler meat will increase to nearly 6500 thousand metric tons by 2010 from the current of 5392.73 thousand metric tons (FAPRI, 2005).



The poultry processing industry has also seen extensive development in the last 30-45 years, to meet the increased demand and has brought the food meat, which was available to limited class of consumer, to the popular, cheap and wholesome meat within everyone's budget (Mulder, 1999). Primary processing of poultry, however, consists of a number of operations, which have microbiological implications. From the farm gate, when birds are transported, to the processing plant, up to chilling and packaging, have all been implicated in some form of microbial cross-contamination. This problem is compounded with the high throughput of modern poultry processing plants, which leaves little room for effectively sanitizing of equipment. Growth of bacteria on meat results in the production of 'off' odours and flavours or an unacceptable appearance to the consumer. This is generally termed microbial spoilage. Microbial spoilage is delayed, but not prevented, by storage of meat at temperatures between -1° to 5°C .

Epidemiological reports from all over the world incriminate poultry meat as a source of outbreaks of human food poisoning which has resulted in great economic loss to nations including the loss of human life. Most of these illnesses have been attributed to cross-contamination with human pathogenic organisms such as *Salmonella* spp, *Escherichia coli*, *Listeria monocytogenes* and *Campylobacter* spp. Several bacterial pathogens have been associated with poultry-borne human illnesses; however, *Salmonella* and *Campylobacter* have been of primary concern. The FAO/WHO (2002) report on foodborne diseases in Europe indicated that about 26% of foods in outbreaks involved poultry and poultry products (including eggs). *Salmonella* serovars are the most frequently reported causal agent of outbreaks in the European region, being responsible for 77.1% of outbreaks. Of these, more than one third were

