

CONTENTS

	Page
Contents	(viii)
List of tables	(xiii)
List of figures	(xiv)
Chapter	
1. Introduction	1
Literature review	2
1. Black tiger shrimp and white shrimp	2
2. Quality changes of shrimp during storage	3
2.1. Degradation of shrimp muscle associated with proteinases	4
2.2. Characteristics of shrimp proteinases	7
2.3. Deterioration of shrimps by microorganisms	8
2.4. Melanosis	9
2.4.1. Polyphenol oxidase (PPO)	11
2.4.2. Inhibition of melanosis	14
2.4.2.1. Reducing agents/Antioxidants	15
2.4.2.2. Acidulants	17
2.4.2.3. Chelators	18
2.4.2.4. Hexylresorcinol	19
3. Modified atmosphere packaging	20
4. Used of MAP for shelf-life extension of fishery products	22
5. Use of MAP for shelf-life extension of shrimps	28
Objectives	30
2. Materials and Methods	31
1. Chemicals and microbial media	31
2. Instruments	31
3. Shrimp samples	32
4. Characterization of shrimp enzymes associated with the quality changes	32

CONTENTS (Continued)

	Page
4.1. Preparation of shrimp enzyme extracts	32
4.2. Enzyme assay	33
4.2.1. Protease activity	33
4.2.2. Collagenase activity	33
4.2.3. PPO activity	33
4.3. characterization of shrimp enzymes	34
4.3.1. pH and temperature profiles	34
4.3.2. Thermal and pH stability	34
4.3.3. Inhibitor study	34
5. Effect of different MAP conditions on melanosis and quality of black tiger shrimp and white shrimp	35
5.1. Sample preparation	35
5.2. Chemical analyses	35
5.2.1. pH determination	35
5.2.2. Determination of TVB and TMA contents	35
5.2.3. Determination of thiobarbituric acid reactive substances	36
5.2.4. Determination of formaldehyde content	36
5.2.5. Determination of TCA soluble peptide content	37
5.3. Physical analyses	37
5.3.1. Determination of shear force	37
5.3.2. Determination of water holding capacity	37
5.3.3. Determination of melanosis	37
5.4 Microbiological analyses	38
5.4.1 Total viable count	38
5.4.2 Lactic acid bacteria count	38
5.5. Enzyme activities	38

CONTENTS (Continued)

	Page
6. Combination effect of melanosis inhibitors and MAP on the quality changes of white shrimp	38
6.1 Effect of phosphate and ascorbic acid pretreatment on quality of white shrimp kept under MAP	39
6.2 Effect of phosphate and 4-hexylresorcinol pretreatment on quality of white shrimp	39
6.2.1 Total viable count	40
6.2.2 Lactic acid bacteria count	40
6.2.3 Determination of coliforms bacteria and <i>E. coli</i>	40
6.2.4 Determination of <i>Salmonella</i>	40
6.2.5 Determination of <i>Staphylococcus aureus</i>	41
6.2.6 Determination of <i>Vibrio parahaemolyticus</i>	41
7. Statistical analysis	41
3. Results and discussion	42
1. Characterization of black tiger shrimp and white shrimp enzymes	42
1.1 Proteases from muscle	42
1.1.1 Temperature and pH profiles	42
1.1.2 Temperature and pH stability	45
1.1.3 Effects of inhibitors	47
1.2 Collagenase from cephalothorax and muscle	48
1.2.1 Temperature and pH profiles	48
1.2.2 Temperature and pH stability.	52
1.2.3 Effects of inhibitors	55
1.3 PPO from cephalothorax	56
1.3.1 Temperature and pH profiles	56
1.3.2 Temperature and pH stability	59
1.3.3 Effects of inhibitors	61

CONTENTS (Continued)

	Page
2. Effect of different MAP conditions on melanosis and quality of black tiger shrimp and white shrimp during refrigerated storage	63
Changes in total viable count	63
Changes in lactic acid bacteria count	66
Changes in pH	68
Changes in total volatile bases and trimethylamine contents	70
Changes in TBARS	75
Changes in TCA-soluble peptide content	78
Changes in formaldehyde	80
Changes in melanosis	82
Changes in water holding capacity	84
Changes in shear force	86
Changes in proteases, collagenase and PPO activity	88
3. Effect of phosphate and ascorbic acid pretreatment on the quality of white shrimp kept under MAP	90
Changes in total viable count	90
Changes in lactic acid bacteria count	91
Changes in pH	92
Changes in total volatile bases and trimethylamine contents	94
Changes in TBARS	96
Changes in TCA-soluble peptide content	97
Changes in formaldehyde	98
Changes in melanosis	100
Changes in water holding capacity	101
Changes in shear force	103
Changes in proteases, collagenase and PPO activity	105

CONTENTS (Continued)

	Page
4. Effect of phosphate and 4-hexylresorsinol pretreatment on quality of white shrimp kept under MAP	108
Changes in microbiological	108
Changes in pH	112
Changes in total volatile bases and trimethylamine contents	113
Changes in TBARS	116
Changes in TCA-soluble peptide content	117
Changes in formaldehyde	119
Changes in melanosis	120
Changes in water holding capacity	122
Changes in shear force	123
Changes in proteases, collagenase and PPO activity	125
4. Conclusions	128
Future works	129
References	130
Appendix	162
Vitae	169

LIST OF TABLES

Table	Page
1. Color scale used to describe the progression of melanosis (black spot) on shrimp	11
2. Representative inhibitors of melanosis	14
3. Shelf-life of fresh fishery products packaged under MA, vacuum or air	24
4. Effect of various inhibitors on the activity of proteases from the muscles of white shrimp and black tiger shrimp	48
5. Effects of various inhibitors on the activity of collagenase from cephalothorax and muscle of white shrimp and black tiger shrimp	56
6. Effect of various inhibitors on the activity of PPO from the cephalothorax of black tiger shrimp and white shrimp	62
7. Changes in coliforms (MPN/g) of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	111

LIST OF FIGURES

Figure	Page
1. Melanosis progression scale of shrimp	11
2. Monophenol oxidase pathway producing the diphenol	12
3. Diphenol oxidase pathway producing the quinones	13
4. Mechanism of prevention of color formation by ascorbic acid	17
5. Temperature profiles of proteases from black tiger shrimp and white shrimp muscles	44
6. pH profiles of proteases from black tiger shrimp and white shrimp muscles	44
7. Thermal stability of proteases from black tiger shrimp and white shrimp muscles	46
8. pH stability of proteases from black tiger shrimp and white shrimp muscles	46
9. Temperature profiles of collagenase from the cephalothorax and muscle of black tiger shrimp and white shrimp	50
10. pH profiles of collagenase from the cephalothorax and muscle of black tiger shrimp and white shrimp	51
11. Thermal stability of collagenase from the cephalothorax and muscle of black tiger shrimp and white shrimp	53
12. pH stability of collagenase from the cephalothorax and muscle of black tiger shrimp and white shrimp	54
13. Temperature profiles of PPO from the cephalothorax of black tiger shrimp and white shrimp	58
14. pH profiles of PPO from the cephalothorax of black tiger shrimp and white shrimp	58
15. Thermal stability of PPO from the cephalothorax of black tiger shrimp and white shrimp	60
16. pH stability of PPO from the cephalothorax of black tiger shrimp and white shrimp	61
17. Changes in TVC (CFU/g) of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	65

LIST OF FIGURES (Continued)

Figure	Page
18. Changes in Lactic acid bacteria (CFU/g) of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	67
19. Changes in pH of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	69
20. Changes in TVB contents of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	73
21. Changes in TMA contents of black tiger and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	74
22. Changes in TBARS of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	77
23. Changes in TCA-soluble peptides of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	79
24. Changes in formaldehyde of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	81
25. Changes in melanosis score of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	83
26. Changes in WHC of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	85
27. Changes in shear force of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	87
28. Changes in proteases, collagenase and PPO activity of black tiger shrimp and white shrimp with and without 0.5 g/l ascorbic acid pretreatment stored under different conditions at 4°C	89
29. Changes in TVC (CFU/g) of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	91

LIST OF FIGURES (Continued)

Figure	Page
30. Changes in lactic acid bacteria (CFU/g) of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	92
31. Changes in pH in white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	93
32. Changes in TVB (A) and TMA (B) contents of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	95
33. Changes in TBARS of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	97
34. Changes in TCA-soluble peptide contents of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	98
35. Changes in formaldehyde of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	99
36. Changes in melanosis score of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	101
37. Changes in WHC of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	103
38. Changes in shear force of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	105

LIST OF FIGURES (Continued)

Figure	Page
39. Changes in proteases, collagenase and PPO activity of white shrimp with and without pretreatment using 2% (w/v) pyrophosphate pretreatment and/ or 5.0 g/l ascorbic acid and stored under different packaging atmospheres at 4°C	107
40. Changes in mesophilic, psychrophilic bacteria and lactic acid bacteria (C) counts (log CFU/g) of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	111
41. Changes in pH of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	113
42. Changes in TVB and TMA contents of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	115
43. Changes in TBARS of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	117
44. Changes in TCA-soluble peptides of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	118
45. Changes in formaldehyde content of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	120
46. Changes in melanosis score of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	122

LIST OF FIGURES (Continued)

Figure	Page
47. Changes water holding capacity of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	123
48. Changes shear force of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	124
49. Changes in proteases, collagenase and PPO activity of whole or decapitated white shrimp with and without 2% pyrophosphate and/ or 0.25% 4-hexylresorcinol pretreatment stored under different conditions at 4°C	127