#### Contents

	Page
Contents	
(9)	
List of Tables	
(11)	
List of Figures	
(12)	
1. Introduction	1
Literature Review	3
1. Tuna / tuna waste	3
2. Fish viscera / proteolytic activity	7
3. Fish sauce	13
4. Microbiology of fish sauce	22
5. Changes during fermentation of fish sauce	24
6. Factors affecting the quality of fish sauce	32
7. Formation and degradation of histamine and other biogenic	37
amines in fish and fish sauce	
Objectives	42
2. Materials and methods	43
1. Materials	43
2. Instruments	44
3. Study on the characterization of proteinase in internal organs	
of skipjack tuna	44

(9)

4.	Study on the effect of storage condition on chemical and	
	microbiological changes of tuna internal organs during storage	47

## **Contents (Continued)**

	Page
5. Study on the effect of storage condition on chemical, physical	
and microbiological changes of tuna internal organs during	
fermentation	51
6. Study on the role of proteinases in fish sauce fermentation of	
skipjack tuna internal organs	56
7. Statistical analysis	57
3. Results and Discussion	58
1. Characterization of proteinases in internal organs of skipjack tuna	58
2. Effect of storage condition on changes of individual tuna internal	
organs	67
3. Effect of storage condition of tuna internal organs on chemical	
and biochemical changes of fish sauce during fermentation	86
4. Effect of calcium chloride and pH on proteolytic activity and	
changes of fish sauce from tuna internal organs with different	
salt levels during fermentation	110
5. Effect of calcium chloride and pH on physical changes of fish	
sauce from tuna internal organs with different salt levels during	
aging	143
4. Conclusion	153

(10)

References	155
Appendix	188
Vitae	204

#### List of Tables

Table	Page
1 Nutritional composition of three species of fish used for fish sauce	34
production.	
2 Distribution of proteinase activity in different internal organs of	
skipjack tuna	62
3 Changes in biogenic amine contents (ppm) in tuna internal organs during	
storage in ice and at room temperature for up to 8 h	79
4 Changes in $\beta$ - phenylethylamine content of fish sauce produced from tuna	
internal organs during fermentation for up to 12 months	98
5 Changes in putrescine content of fish sauce produced from tuna internal	
organs during fermentation for up to 12 months	99
6 Changes in cadavarine content of fish sauce produced from tuna internal	
organs during fermentation for up to 12 months	99
7 Changes in histamine content of fish sauce produced from tuna internal	
organs during fermentation for up to 12 months	100
8 Changes in tyramine content of fish sauce produced from tuna internal	
organs during fermentation for up to 12 months	100
9 Changes in spermidine content of fish sauce produced from tuna internal	

# List of Figures

Figure	Page
1 Tuna species	6
2 Temperature profiles of individual and pooled internal organs from	
skipjack tuna	61
3 pH profiles of individual and pooled internal organs from skipjack tuna	61
4 Effect of NaCl concentrations on proteinase activity of different internal	
organs from skipjack tuna	64
5 Changes in proteolytic activity of different internal organs from	
skipjack tuna during fermentation	66
6 Changes in TCA-soluble peptide of individual internal organs during	
storage for up to 8 h	68
7 Changes in pH of individual internal organs during storage for up to	
8 h	70
8 Changes in TVB content of individual internal organs during storage	
for up to 8 h	73
9 Changes in TMA content of individual internal organs during storage	
for up to 8 h	74
10 Changes in histamine content of individual organ during storage	

for up to 8 h	76
11 Changes in microbial count of individual internal organs during	
storage for up to 8 h	81

Figure	page
12 Changes in halophilic bacteria of individual internal organs during	
storage for up to 8 h	83
13 Changes in proteolytic bacteria of individual internal organs during	
storage for up to 8 h	85
14 Changes in pH of fish sauce from different quality tuna internal organs	
during fermentation of 12 months	86
15 Changes in TVB and TMA contents of fish sauce from different quality	
tuna internal organs during fermentation of 12 months	88
16 Changes in total nitrogen content of fish sauce from different quality	
tuna internal organs during fermentation of 12 months	90
17 Changes in formol nitrogen content of fish sauce from different quality	
tuna internal organs during fermentation of 12 months	91
18 Changes in ammoniacal nitrogen content of fish sauce from different	
quality tuna internal organs during fermentation of 12 months	92
19 Changes in amino nitrogen content of fish sauce from different quality tuna	
internal organs during fermentation of 12 months	93
20 Changes in salt contents of fish sauce from different quality tuna internal	

organs during fermentation of 12 months	94
21 Changes in histamine content of fish sauce from different quality tuna	
internal organs during fermentation of 12 months	96

Figure	Page
22 Changes in fluorescence intensity of fish sauce from different quality tuna	
internal organs during fermentation of 12 months	102
23 Changes in $A_{294}$ of fish sauce from different quality tuna internal organs	
during fermentation of 12 months	104
24 Changes in $A_{420}$ of fish sauce from different quality tuna internal organs	
during fermentation of 12 months	105
25 Changes in color values of fish sauce from different quality tuna internal	
organs during fermentation of 12 months	107
26 Changes in total viable count, halophilic bacteria and proteolytic bacteria	
count of fish sauce from different quality tuna internal organs during	
fermentation of 12 months	109
27 Changes in proteolytic activity of fish sauce from tuna internal organs as	
affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	113
28 Changes in total nitrogen content of fish sauce from tuna internal organs as	
affected by salt, CaCl2 and pH adjustment during fermentation	116
29 Changes in formal nitrogen content of fish sauce from tuna internal organs	
as affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	118

(14)

30	Changes in ammonia nitrogen content of fish sauce from tuna internal	
	organs as affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	121
31	Changes in amino nitrogen content of fish sauce from tuna internal organs	
	as affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	123
	as affected by suit, eacing and pir adjustment during rementation	

Figure	page
32 Changes in pH of fish sauce from tuna internal organs as affected by	
salt, CaCl <sub>2</sub> and pH adjustment during fermentation	125
33 Changes in TVB content of fish sauce from tuna internal organs as	
affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	128
34 Changes in TMA content of fish sauce from tuna internal organs as	
affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	129
35 Changes in $A_{294}$ of fish sauce from tuna internal organs as affected	
by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	131
36 Changes in fluorescence intensity of fish sauce from tuna internal organs	
as affected by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	134
37 Changes in $A_{420}$ of fish sauce from tuna internal organs as affected	
by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	137
38 Changes in L*- value of fish sauce from tuna internal organs as affected	
by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	140
39 Changes in a*- value of fish sauce from tuna internal organs as affected	
by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	141
40 Changes in b*- value of fish sauce from tuna internal organs as affected	

(15)

by salt, CaCl <sub>2</sub> and pH adjustment during fermentation	142
41 Changes in $A_{294}$ of fish sauce from tuna internal organs as affected	
by salt, CaCl <sub>2</sub> and pH adjustment during aging	144

Figure	Page
42 Changes in fluorescence intensity of fish sauce from tuna internal	
organs as affected by salt, CaCl <sub>2</sub> and pH adjustment during aging	146
43 Changes in $A_{420}$ of fish sauce from tuna internal organs as affected by	
salt, CaCl <sub>2</sub> and pH adjustment during aging	148
44 Changes in L*-value of fish sauce from tuna internal organs as affected by	
salt, CaCl <sub>2</sub> and pH adjustment during aging	150
45 Changes in a*-value of fish sauce from tuna internal organs as affected by	
salt, CaCl <sub>2</sub> and pH adjustment during aging	151
46 Changes in b*-value of fish sauce from tuna internal organs as affected by	
salt, CaCl <sub>2</sub> and pH adjustment during aging	152