

TABLE OF CONTENTS

	Page
TABLE OF CONTENTS.....	i
LIST OF TABLES.....	iii
LIST OF FIGURES.....	v
INTRODUCTION.....	1
OBJECTIVES.....	3
LITERATURE REVIEWS.....	4
Carbofuran.....	4
Persistence of Pesticides in Soil.....	6
Fate of Carbofuran in Soil.....	9
Pesticide-Degrading Bacteria and Their Metabolism	10
MATERIALS AND METHODS.....	15
Materials.....	15
Methods.....	15
RESULT AND DISCUSSIONS.....	19
Properties of Soil Samples.....	19
Number of Culturable Microorganisms from Carbofuran Treated Soils.....	20
Isolation and Characterization of Carbofuran Degrading Microorganisms.....	22
Carbofuran Residue Determination by TLC.....	43
Carbofuran Residue Determination by HPLC.....	44
Degradation Abilities of 66 isolates.....	46
Effect of pH, Light and Inoculum Size on Degradation of Carbofuran in Minimal Medium.....	54
Effect of pH on Carbofuran Degradation in Minimal Medium.....	55
Effect of Light on Carbofuran Degradation in Minimal Medium.....	56
Effect of Inoculum Size on Carbofuran Degradation in Minimal Medium.....	57
Metabolism Metabolites Produced from Carbofuran Degradation Process.....	58

TABLE OF CONTENTS (CONTINUED)

	Page
CONCLUSION.....	62
LITERATURE CITED.....	64
APPENDIX.....	70
Appendix A.....	71
Appendix B.....	74

LIST OF TABLES

Table		Page
1	The properties of 2 soil samples collected from Bangsapan, Chaikasem district, Prachaupkhirikhan province.....	19
2	Effect of carbofuran concentration at 1X and 2X to the number of culturable microorganisms of soil A and B compared with control.....	20
3	Grouping of carbofuran degrading <i>Bacillus</i> sp. by morphological properties.....	23
4	List of <i>Bacillus</i> isolates that belonged in group IA classified as <i>B. megaterium</i> and <i>B. thuringiensis</i>	25
5	Biochemical and physiological characteristics of isolate P001, P002, P007, P008, P009, P011, P021, P028, P029, K002, K003, K004, K005, K009, K014, K018 and K024 compared with characteristics of <i>B. megaterium</i>	28
6	Biochemical and physiological characteristics of isolate P006, P010, P012, P023, P024, P026, P027, P030, P031, P033, K006, K011, K015, K026, K031, K032 and K033 compared with characteristics of <i>B. thuringiensis</i>	29
7	Isolate of <i>Bacillus</i> sp. belonged in group IB.....	31
8	Biochemical and physiological characteristics of isolate P013, P014, P015, P016, P017, P018, P019, P020, K008, K013, K016, K019, K021, K025 and K027 compared with <i>B. amyloliquefaciens</i>	33
9	Biochemical and physiological characteristics of unidentified isolate K010 and K029.....	35
10	List of <i>Bacillus</i> isolates belonged in group II.....	36
11	Biochemical and physiological characteristics of isolate P003, P004, P005, K007, K012 and K023 compared with <i>B. brevis</i>	38
12	Biochemical and physiological characteristics of isolate K001, K017 and K020 compared with <i>B. circulans</i>	40
13	Biochemical and physiological characteristics of isolate K022 compared with <i>B. sphaericus</i>	42

LIST OF TABLES (CONTINUED)

Table	Page	
14	Twenty eight isolates which capable to degrade carbofuran more than 50% within 7 days.....	53

Appendix Table

B1	Carbofuran residual percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IA (<i>B. megaterium</i>).....	74
B2	Carbofuran residual percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IA (<i>B. megaterium</i>).....	75
B3	Carbofuran residual percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IA (<i>B. thuringensis</i>).....	76
B4	Carbofuran residual percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IA(<i>B. thuringensis</i>).....	77
B5	Carbofuran residual percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IB.....	78
B6	Carbofuran residual percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IB.....	79
B7	Carbofuran residual percentage at 3, 5 and 7 day of the isolates that belonged to genus <i>Bacillus</i> in group II and III.....	80

LIST OF FIGURES

Figure		Page
1	The molecular structure of carbofuran.....	4
2	The interactions influencing the fate of the pesticide in soil.....	8
3	Some microbial reaction that effect <i>N</i> -methylcarbamate.....	10
4	The structure of metabolites produced during the carbofuran degradation process.....	12
5	<i>B. megaterium</i> isolate.....	26
6	<i>B. thuringiensis</i> isolate.....	27
7	<i>B. amyloliquefaciens</i> isolate.....	32
8	Isolate K010, representing of <i>Bacillus</i> group IB, grown on NA not more than 24 hours showed vegetative cells, sporangium and cell arrangement.....	34
9	Isolate K029, representing of <i>Bacillus</i> group IB, grown on NA not more than 24 hours showed vegetative cells, sporangium and cell arrangement.....	34
10	<i>Bacillus</i> sp. closely to <i>B. brevis</i>	37
11	Unidentified <i>Bacillus</i> sp. (closed to <i>B. circulans</i>).....	39
12	Isolate K022, representing of <i>Bacillus</i> group III identified as <i>B. sphaericus</i> , grown on NA not more than 24 hours showed vegetative cells, sporangium and cell arrangement.....	41
13	Chromatogram of standard carbofuran which had retention time at 26.812 minutes.....	44
14	Standard curve of carbofuran compared between peak area and concentration.....	45
15	Relative degradation ability percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IA identified as or closed to <i>B. megaterium</i> compared with control.....	46

LIST OF FIGURES (CONTINUED)

Figure		Page
16	Relative degradation ability percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IA identified as <i>B. megaterium</i> compared with control.....	47
17	Relative degradation ability percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IA identified as or closed to <i>B. thuringensis</i> compared with control.....	48
18	Relative degradation ability percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IA identified as or closed to <i>B. thuringensis</i> compared with control.....	49
19	Relative degradation ability percentage at 3, 5 and 7 day of “P” isolates that belonged to genus <i>Bacillus</i> in group IB identified as <i>B. amyloliquefaciens</i> compared with control.....	50
20	Relative degradation ability percentage at 3, 5 and 7 day of “K” isolates that belonged to genus <i>Bacillus</i> in group IB (<i>B. amyloliquefaciens</i> and <i>Bacillus</i> sp...)	51
21	Relative degradation ability percentage at 3, 5 and 7 day of the isolates that belonged to genus <i>Bacillus</i> in group II and group III.....	52
22	Effect of pH 5 and 8 on carbofuran degradation in minimal medium inoculated with isolate P004, P027, K025, K031 and control (no inoculation) at day 5.....	55
23	Effect of light on carbofuran degradation in minimal medium inoculated with isolate P004, P027, K025, K031 and control (no inoculation) at day 5.....	56
24	Effect of inoculum size on carbofuran degradation in minimal medium inoculated with isolate P004, P027, K025, K031 and control (no inoculation) at day 5.....	57
25	The chromatogram of unknown substances in control sample at day 7.....	58
26	The chromatogram of unknown substances in P027 sample at day 7.....	59

LIST OF FIGURES (CONTINUED)

Figure		Page
27	The chromatogram of unknown substances in K004 sample at day 7.....	59
28	Structure formula of 1, 2-benzenedicarboxylic acid.....	60