CONTENTS		PAGE
ABSTRACT		ii
ACKNOWLEDGEMENT		v
LIST OF TABLE	E S	vi
LIST OF FIGURES		vii
DEDICATION		ix
CHAPTER 1	INTRODUCTION	1
CHAPTER 2	LITERATURE REVIEW	4
2.1	Salmonella as pathogen	4
2.2	Salmonella Typhi	5
2.3	Prevalence of typhoid fever	6
2.4	Pathogenicity and clinical features	7
2.5	Diagnosis and detection of typhoid fever	9
2.6	PCR based diagnosis of typhoid fever	11
CHAPTER 3	MATERIALS AND METHODS	13
3.1	Chemicals and biological	13
	3.1.1 Bacterial Strains	13
	3.1.2 Plasmids	13
	3.1.3 PCR Primers	14
	3.1.4 Enzymes	15
3.2	Molecular Techniques	15
	3.2.1 Genomic DNA extraction	15

	3.2.2 Plasmid DNA extractions	16
	3.2.3 PCR amplifications of targeted DNA sequence	17
	3.2.4 Rapid screening for inserts by colony PCR	17
	3.2.5 Restriction digestion of DNA fragment	18
	3.2.6 Gel electrophoresis	18
	3.2.7 Ligation reaction of DNA fragment to vector	19
	3.2.8 Preparation of Escherichia coli competent cells	19
	3.2.9 Transformation of DNA to competent Escherichia	
	coli cells	19
	3.2.10 Induction of protein expression with IPTG	20
	3.2.11 Protein extraction with NP40 buffer	20
	3.2.12 Periplasmic protein extraction	21
	3.2.13 Bradford assay for protein quantification	21
	3.2.14 Colony blotting of transformed <i>Escherichia coli</i>	
	cells	21
	3.2.15 Dot-blot assay	22
	3.2.16 ELISA assay	23
CHAPTER 4	RESULTS	24
4.1	Characterization of specific DNA marker of Salmonella	
	Typhi	24
	4.1.1 Construction of an expression system for specific	
	DNA marker of Salmonella Typhi	24
	4.1.2 PCR amplification of the Salmonella Typhi DNA	
	marker ST332	25
	4.1.3 Immunodetection of Salmonella Typhi specific	

	DNA marker as a hypothetical protein	35
4.2	Screening of specific Salmonella Typhi antigen	37
	4.2.1 Construction of library clones of Salmonella Typhi	
	genomic DNA	37
	4.2.2 Colony immunoscreening of recombinant	
	NovaBlue (Escherichia coli DE3) clones	41
	4.2.3 Immunodetection of the putative reactive clones	43
	4.2.4 Elisa immune detection of potential clones	44
CHAPTER 5	DISCUSSIONS	49
5.1	Characterization of specific DNA marker of Salmonella	
	Typhi	49
5.2	Epitope mapping of Salmonella Typhi	55
CHAPTER 6 CONCLUSION AND FUTURE WORKS		57
REFERENCES		59
APPENDICES		67