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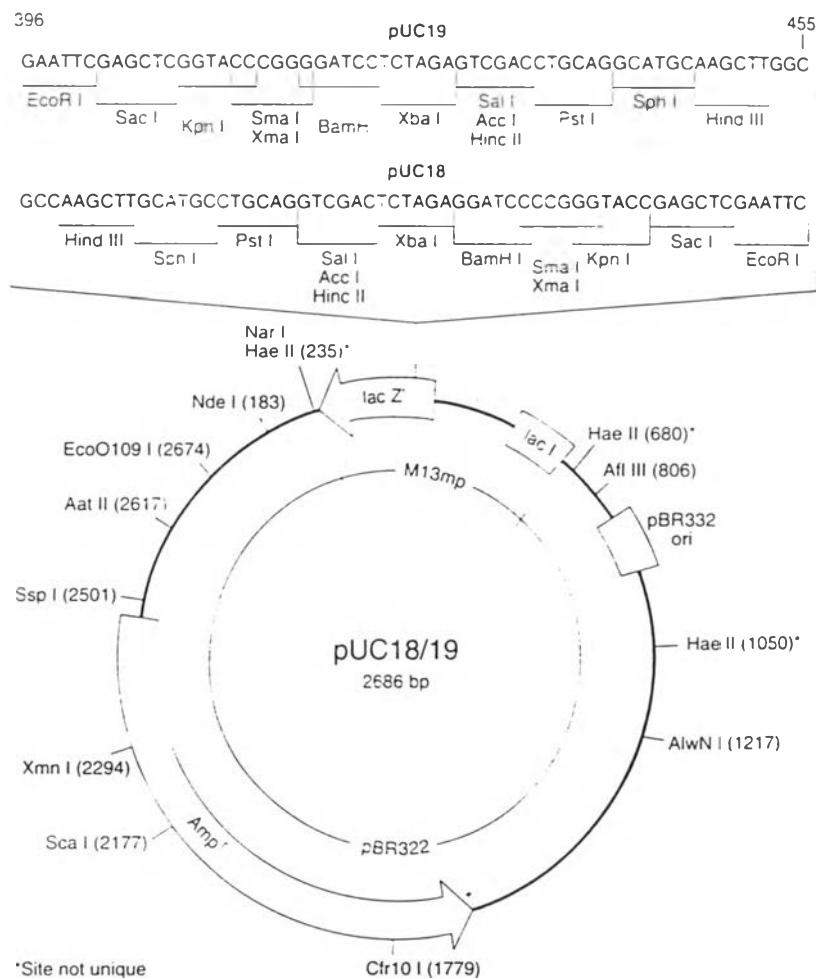
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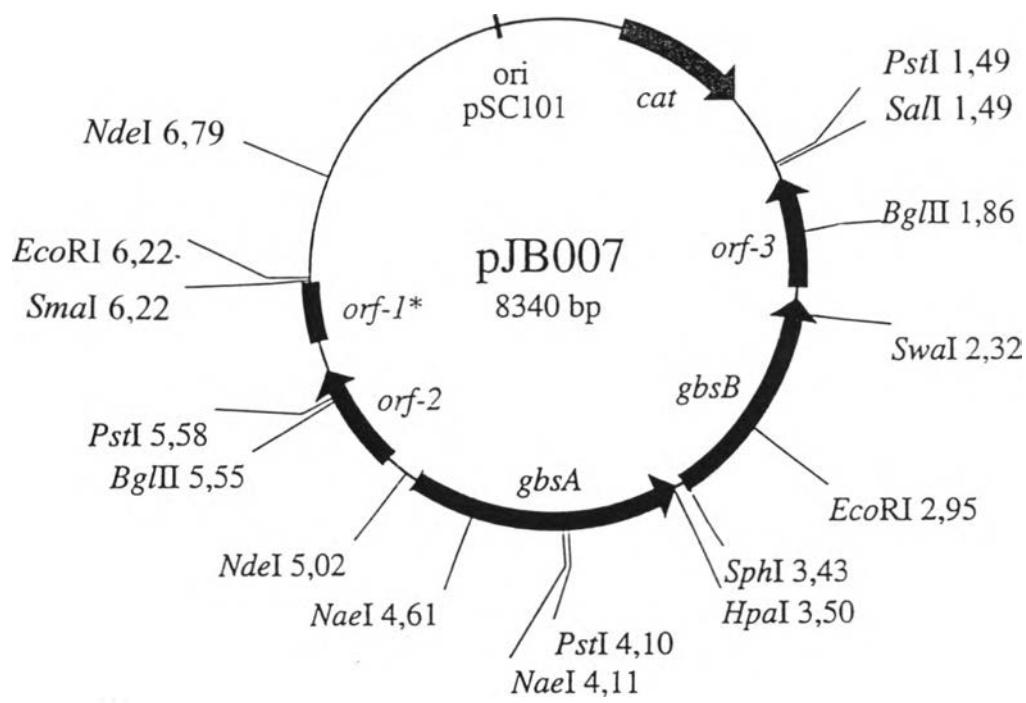
## APPENDIX 1

### Restriction map of pUC18



## APPENDIX 2

## Restriction map of pJB007



### APPENDIX 3

#### Media Preparation

##### **Turk Island Salt solution + modified BG<sub>11</sub> medium**

###### 1. Preparation of Turk Island Salt solution

Stock solution A :	KCl	33.3	g
	MgCl <sub>2</sub> .6H <sub>2</sub> O	275.0	g
	CaCl <sub>2</sub> .2H <sub>2</sub> O	73.3	g

and made up to 5 liters with distilled water

Stock solution B :	MgSO <sub>4</sub> .7H <sub>2</sub> O	374	g
and made up to 5 liters with distilled water			

###### 2. Composition of modified BG<sub>11</sub> medium (BG<sub>11</sub> medium + NaNO<sub>3</sub> solution)

150	g/l	NaNO <sub>3</sub>	50	ml
32	g/l	KH <sub>2</sub> PO <sub>4</sub>	5	ml
60	g/l	MgSO <sub>4</sub> .7H <sub>2</sub> O	5	ml
28.8	g/l	CaCl <sub>2</sub>	5	ml
16	g/l	Na <sub>2</sub> CO <sub>3</sub>	5	ml
4.8	g/l	citric acid	5	ml
0.8	g/l	EDTA.Na <sub>2</sub>	5	ml
4.8	g/l	MgSO <sub>4</sub> .7H <sub>2</sub> O	5	ml

\* Trace element A<sub>s</sub> solution + Co contained the following component in gram per liter ; H<sub>4</sub>BO<sub>4</sub> (2.86), ZnSO<sub>4</sub>.7H<sub>2</sub>O (0.22), CuSO<sub>4</sub>.5H<sub>2</sub>O (0.08), MnCl<sub>4</sub>.4H<sub>2</sub>O (1.81), Na<sub>2</sub>Mo<sub>4</sub>.2H<sub>2</sub>O (0.39) and Co(NO<sub>3</sub>)<sub>2</sub>.6H<sub>2</sub>O (0.049).

Culture medium of *A. halophytica* was prepared by adding all solution of item 2 at indicated volume to 500 ml of Stock solution A and 500 ml of Stock solution B. To this mixture 140.8 g NaCl was added and adjusted pH to 7.6 by slowly adding 2 M NaOH then adjusted the final volume to 5 liters with distilled water. The medium was sterilized by autoclaving at 151 lb/in<sup>2</sup> for 15 minutes.

**LB medium**

per liter

tryptone	10	g
yeast extract	5	g
NaCl	10	g

**M63 medium**

10X M63 stock, per liter

KH <sub>2</sub> PO <sub>4</sub>	30	g
K <sub>2</sub> HPO <sub>4</sub>	70	g
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	20	g
FeSO <sub>4</sub>	5	mg

Before use, dilute concentrated media to 1X with sterile water and add the following sterile solutions, per liter :

1	ml	1 M MgSO <sub>4</sub> .7H <sub>2</sub> O
10	ml	20% glucose
0.1	ml	0.5% vitamin B <sub>1</sub> (thiamine)
5	ml	20% casamino acids

## APPENDIX 4

Sequence comparison of the genes coded for betaine aldehyde dehydrogenase from bacteria: *E. coli* and *B. subtilis*. Identical nucleotides are marked by asterisks.

<i>E. coli</i>	GATGGCAAACCTTCTTCAGCTCCGTCAAGGTGTGACCAATGGCACCCCGC'CTTCGTTCC
<i>B. subtilis</i>	AAACGCTGTATTTTCCACGCCGCCAAGTATGCTCTGCAGGTTCCCGCT'GCTTGGTGA *
<i>E. coli</i>	GGC-GAAATGCAAAGCCGCATTGAGCAGAAAATTCTGGCGCGCGTTGAG'CGATTGCG
<i>B. subtilis</i>	GGATGCCATTCA-T-GATCAATTGGCGGAGCTGGTAAACGGGAAAA'GCATAAAAC *
<i>E. coli</i>	CGGGCAGCTTTGATCGCAGAAACTAACCTCGGCCGCTGGTCAGCTC'CGCATCGCG
<i>B. subtilis</i>	TCGAAACGGTTTCATGCTGAGACAGAAAGCGGTCCGCTTATTCGGCG'AGCACAGGG *
<i>E. coli</i>	ATAACGTGCTGCCTATATGCCAAAGGCAAAGAGGAAGGGCGCGCGTA'TGTGCGGCG
<i>B. subtilis</i>	CAAAGGTAGAAAATATGTAGAGATCGGAATAGAGGAAGGGCGGAAGCTG'AGACAGGG *
<i>E. coli</i>	GCGATGTACTGAAAGGCGATG-GCTTCGATAACGGCGCATGGGTTGCACCGACAGTGTTC
<i>B. subtilis</i>	GCAAACGCCCGGAAGATCCTGAGCTTC-AAAACGGCTTTCTATGAACC'ACTATTTTC *
<i>E. coli</i>	ACCGATTGCAGCGACGATATGACCATCGTGCCTGAAGAGATCTCGGCCGCACTGATGTCC
<i>B. subtilis</i>	TCAAACGTAAATTCTGACATGAGAATCGTCAAGGAAGAGGTTTCGGTCC'GTATTGACA *
<i>E. coli</i>	ATTCTGACCTACGAGTCGAAGACGAAGTCATTGCCGCGCTAACGATAC'GACTACGGC
<i>B. subtilis</i>	GTCGAAACCTTCAGCTCTGAAGAAGAGGTAATCGAGCTTGCAGAACATCTATGGC *
<i>E. coli</i>	CTGGCGCGGGCATCGTGCAGCGGGACTT-GAACCGCGCATCGCGTC'TCATCAGCT
<i>B. subtilis</i>	TTGGCTGGAGCGGTATGGTCAAAAGACATTGAAAAGTGCG-AACGGT-A'CGCCCGCT *
<i>E. coli</i>	GGA-AGCGGGTATTGCTGGATCAACACCTGGGCGAATCCCAGAG'TGCCCGTTG
<i>B. subtilis</i>	TGAGAATGGAACCGTTGGATCAACGATTTCATCGTACTTGCACAA'CGCCATGGG *
<i>E. coli</i>	GCGGCTACAAACACTCCGCATTGGTCGCGAGAACGGCGTGATGACGCTC'AGAGTTACA
<i>B. subtilis</i>	GCGGATATAAGCAATCGGGTTCGGACGCGAGCTGGAAAATAGGCCTT'AGAAATACA *
<i>E. coli</i>	CCCAGGTGAAGTCATCCAGGTGAGATGGCTAAATTCCAGTCCATAT--TCTAA--
<i>B. subtilis</i>	CAGAAGTCAAA-CATGTATACCGCAATACAAAACGGCAGCGGTTAACT'GTTTAATTC *
<i>E. coli</i>	-----
<i>B. subtilis</i>	ATAA

## APPENDIX 5

Sequence comparison of the genes coded for betaine aldehyde dehydrogenase from bacteria: *E. coli* and *R. meliloti*. Identical nucleotides are marked by asterisks.

<i>E. coli</i>	---ATGTCCCGAATGGCAGAACAGCAGCTTATATACATGGGGTTATAC	'TCGCCACC
<i>R. meliloti</i>	ATGAGAGCCAACCGAAAGCCTCGCA-CTTCATCGAC - GGCATAATGT	'GAGGACGCC
	***** *	***
<i>E. coli</i>	AGCGGTCGCACCTTCGAGACCATTAACCCGCCAACGTAACGTGCTGGC	'ACCGTGCAG
<i>R. meliloti</i>	GCCGGCACGGTGATCGAGAGCATCTATCCGGGACCGGCAAATCATCGC	'CGGCTCCAT
	*** *	**
<i>E. coli</i>	GCCGCCGGGCGCGAGGATGTCGATCGGCCGTGAAAAGGCCAGCAGGG	'CAAAAATC
<i>R. meliloti</i>	GCGCAACGCCCTGGGATCGTCGAGAAGGCGATCGCCGCGGCCAGCGCCG	'CAGCCGGAA
	***** *	*
<i>E. coli</i>	TGGGCGTCGATGACCGCCATGGAGCGCTCGGTATTCTGCGTCGGCCGT	'GATATTCTG
<i>R. meliloti</i>	TGGGCGCGATGACCCGACGGCGCGGCCATCCTGAAGCGGGCCG	'GAGCTCATG
	***** *	**
<i>E. coli</i>	CGTGAACGCAATGACGAACTCGAAAATGGAAACCCCTCGACACCGGAA	'GCATATTG-
<i>R. meliloti</i>	CGCCAGCTCAACCGCAGCTTCGAACTCGAAACGCTCGACACCGGAA	'GCCGATCCA
	*** *	*
<i>E. coli</i>	GGAAACCTCAACCGTCGATATCGTTACCGGTGCGGACGTGCTGGAGTACT	'CGCCGGGCT
<i>R. meliloti</i>	GGAAACCATCGTCGCCGACCCGACGTCCGGCGCCGACAGTTCTGAATTCT	'CGGC GGCGT
	***** *	*
<i>E. coli</i>	-GATCCC GGCGC - TGGAAAGGCAGCCAGATCCGTTGCGTAAACGTCT	'TGTGTATAC
<i>R. meliloti</i>	CGCGCCCGCCGCACTAACGGCATTATATCCGCTCGGC - GCGAC-T	'CGCCTATAC
	* *	*****
<i>E. coli</i>	CCGGCCCGAACCGTGGGCTAGTGGCAGGGATTGGCGCATGGAAC	'GATCCAGAT
<i>R. meliloti</i>	GAAGCGGGTGCCTCGCGCTCGCGCATCGCGCTGGAACTATC	'GCAGCAGAT
	*** *	*****
<i>E. coli</i>	TGCCCTGTGAAATCCGCCCCGGCGCTGGCGCAGGCAACGCAATGATT	'CAAACCGAG
<i>R. meliloti</i>	CGCCTGCTGGAAGGGTGCCTCGCGCTCGCGCCGAAATGCGATGGTGT	'CAAGCCTTC
	*** *	*****
<i>E. coli</i>	CGAAGTTACCCGCTTACCGCGTAAAGCTGGCTGAAATTACAGCGAAG	'GGGCCTGCC
<i>R. meliloti</i>	GGAAAACACCCCGCTCGCGCGCTTAAGATCGCCGAAATCTTATCGAAG	'GGGTCTGCC
	*** *	*****
<i>E. coli</i>	GGACGGCGTATTTAACGTGTTGCCGGCGTGGCGCGAGACGGGCAAT	'TCTGACCGA
<i>R. meliloti</i>	GAAGGGCTGTTCAACGTCTCAGGGCGACCCCGC - GACGGGCCCCG	'CCTCGTCAA
	* *	** * *
<i>E. coli</i>	GCATCCGGGATTGCCAAAGTGTCAATTACCGCGGTGTCGGCAGCGGA	'AAAAGTGAT
<i>R. meliloti</i>	CCATCCGGACGTGCCAACGGTGTGCGCTCACC CGCTCGGCCAGGGCA	'AAAAGTCGC
	***** *	*****
<i>E. coli</i>	GGCTAACTCGCGGCCTTCCCTGAAAGAAGTGACCATGGAAC	'TAAATCACC
<i>R. meliloti</i>	GGCGCCGCCGGCGCCG --- AACTCAAGCACGTACCATGGAGCTGGCG	'CAAGTCGCC
	** *	*****
<i>E. coli</i>	GCTGATCGTTTCGATGATGCGGATCTCGATCTCGCCGCCGATATGCCA	'GATGGCAA
<i>R. meliloti</i>	GCTGATCGTCTCGACGATGCCGATCTCGAAAGCGCGATCGCGGCCA	'GCTCGCAA
	***** *	***** * * * *

<i>E. coli</i>	CTTCTTCAGCTCCGGTCAGGTGTGACCAATGGCACCCCGCTTCTGTC	:GGCGAAATG		
<i>R. meliloti</i>	TTTCTATTGACCGGCCAGGTCTGCTCCAACGGCACGCGCTCTCGTGC	:AAGGAAGAT		
<b>*****</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	CAAAGCCGCATTTGAGCAGAAAATTCT	-GGCGCGCTTGAGCGATTCGC	:CGGGCGACG	
<i>R. meliloti</i>	CAA-GGAGCCTTCTCGCCCGCCTCAAGGAACGCACCGAACCGATCGTC	:TCGGCGACC		
<b>***</b> <b>*</b> <b>***</b> <b>*</b> <b>*</b> <b>**</b> <b>***</b> <b>*</b> <b>***</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	TTTTCGATCCGCAA	ACTA	ACTTCGGCCCGCTGGTCAGCTCCGCATCGC	:ATAACGTGC
<i>R. meliloti</i>	CCCTGGACGAGGCCACGCAGCTCGGTCCGATGGTCTCGGGCGCGAACGC	:ACAAGGTCT		
<b>***</b> <b>*</b> <b>**</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b>				
<i>E. coli</i>	TGCGCTATATCGCCAA	AGGCAAAAGAGGAAGGGCGCGCGTACTGTGCGGC	:KGCGATGTAC	
<i>R. meliloti</i>	TCTCCTATATCGGGAAAGGCAAGGCGGAAGGC	GGCGCCCGCTCGTCACCGGC	:GCGGCATTC	
<b>*</b> <b>*****</b> <b>***</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*</b>				
<i>E. coli</i>	TGAAAGGCATGGCTTCGATAACGGCGCATGGGTTGCACCGACAGTGTTC	:ACCGATTGCA		
<i>R. meliloti</i>	CCAATA--ATGTGAGCGGCAGGGCACCTATATCCAGCCACCGTCTTC	:CCGACGACA		
<b>**</b> <b>***</b> <b>*</b> <b>**</b> <b>*</b> <b>*</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	GCGACGATATGACCATCGTGCCTGAAAGAGATCTCGGGCCAGTGATGTCC	:TTCTGACCT		
<i>R. meliloti</i>	CCGACGGGATGACGATCGCGCGAAGAAATCTCGGACCGTCATGTG	:TGCTCGACT		
<b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	ACGAGTCGGAAGACGAAGTCATTGCCGCGTAACGATAACGACTACGGC	:TGGCGCCGG		
<i>R. meliloti</i>	TCGACGACGAGGTGGAAGTCATCGCACGCGCCAACGCCACCGAACCGG	:TTTCGGCCG		
<b>***</b> <b>*</b> <b>**</b> <b>*****</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	GCATCGTACAGCGGACCTGAAACCGCGCGCATCGCGTCATT	CATCAGCTG	:AAGCGGGTA	
<i>R. meliloti</i>	GCGTCTCACCGCGGACCTCACCGCGCCACCGGGTCGCCACCGCTC	:AAGCCGGCA		
<b>***</b> <b>*</b> <b>**</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	TTTGTGGATCAACACCTGGGGCAATCCCCGGCAGAGATGCCC	GTTGGC	:GCTACAAAC	
<i>R. meliloti</i>	CGCTCTGGATCAACACCTACAATCTCTGCCGGTAGAGATCCC	TTTGGC	:GGTCCAAGC	
<b>*****</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	ACTCCGGCATTGGTCGCGAGAACGGCGTGATGACGCTCCAGAGTTACACC	:AGGTGAAGT		
<i>R. meliloti</i>	AATCGGGTTTCGGCGGGAGAACGGTCGCGCGCTAACCACTATACC	:AGCTCAAAGA		
<b>*</b> <b>**</b> <b>*</b> <b>**</b> <b>***</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b> <b>*</b> <b>*****</b>				
<i>E. coli</i>	CCATCCAGGTTGAGATGGCTAAATTCCAGTCATATTCTAA			
<i>R. meliloti</i>	CCGTCTATGTCGGCATGGGGCCGGTCGAGGC	GGCGCGTATTGA		
<b>***</b> <b>*</b> <b>**</b> <b>*</b> <b>*****</b> <b>*</b> <b>**</b> <b>*</b> <b>*</b> <b>*</b>				

## APPENDIX 6

Sequence comparison of the genes coded for betaine aldehyde dehydrogenase between *E. coli* and *R. meliloti*. Identical nucleotides are marked by asterisks.



## BIOGRAPHY

Miss Nadthanon Phusi was born on June 20, 1973 in Nakornphathom, Thailand. She graduated with a Bachelor of Science Degree in Biology from Burapha University in 1994. She has further studied for the Master of Science Degree in Biotechnology Program, Chulalongkorn University since 1995.

