

APPENDIX A

Preparation of stock solution for 0% and 100% denaturant solutions.

Percentage of denaturant solution	0%	100%
40% Acrylamide / bis solution, 37:5:1	4.0 ml	4.0 ml
50X Tris Acetate EDTA (TAE) buffer	0.4 ml	0.4 ml
Formamide	-	8.0 ml
Urea	-	8.4 ml
Sterile Milli-Q water top up to	20.0 ml	20.0 ml

APPENDIX B

Preparation of 25% and 65% denaturant solutions.

Stock of denaturant solution	25%	65%
0%	13.5 ml	6.3 ml
100%	4.5 ml	11.7 ml

APPENDIX C

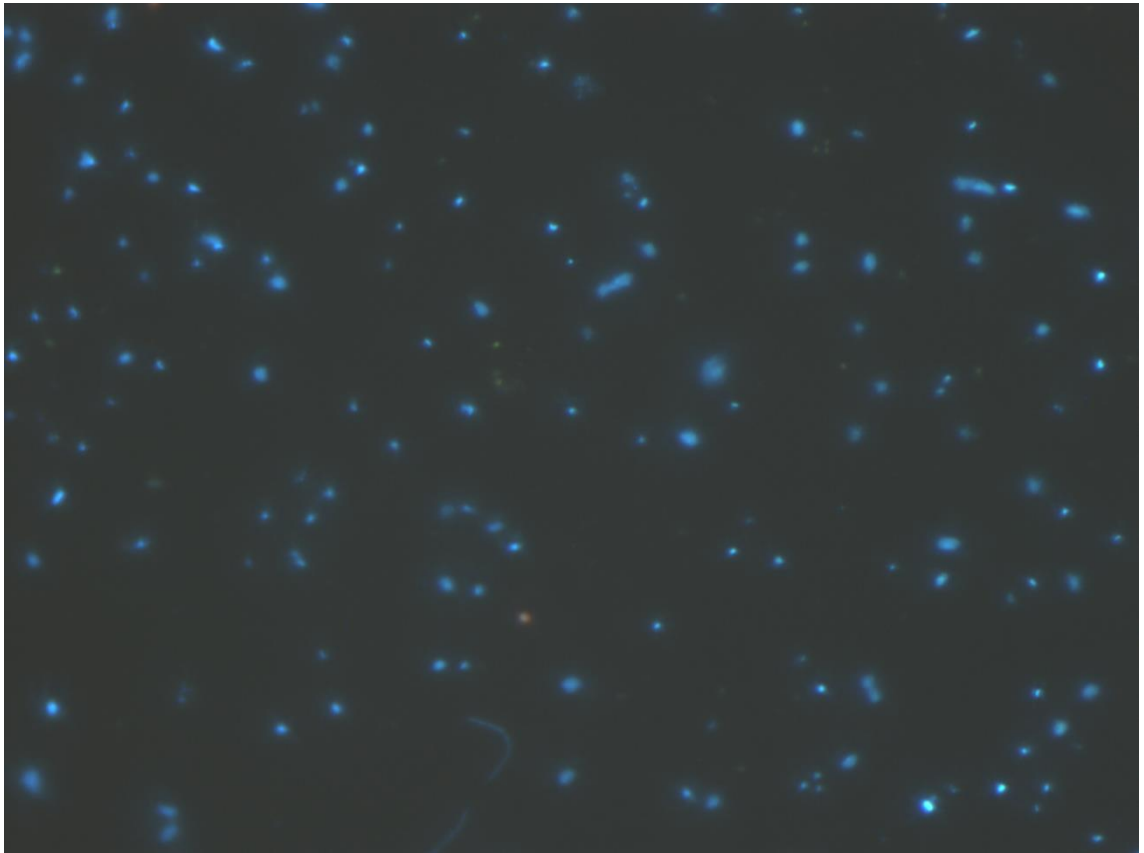


Photo of DAPI stained bacteria which viewed under epifluorescence microscope with a U-MWU filter cassette (excitor 330-385 nm, dichroic mirror 400 nm, barrier 420 nm) for determination of bacterial abundance.

APPENDIX D

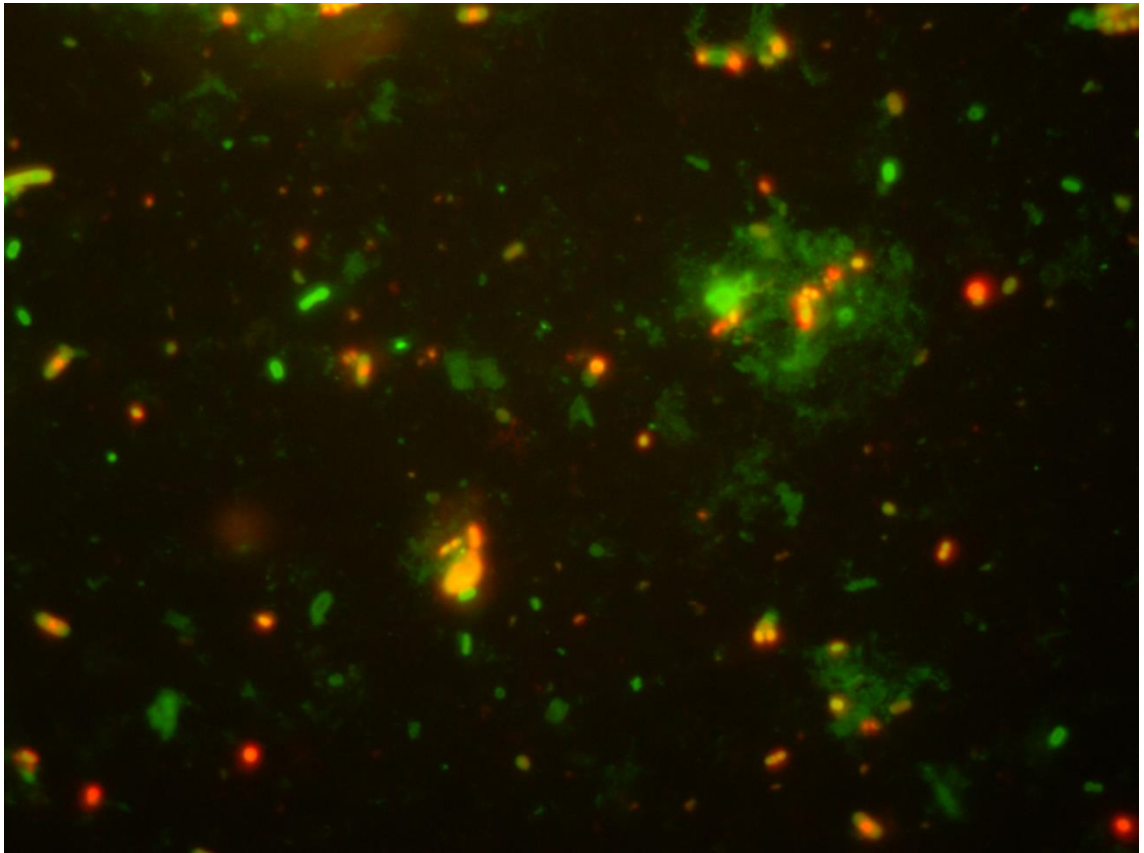
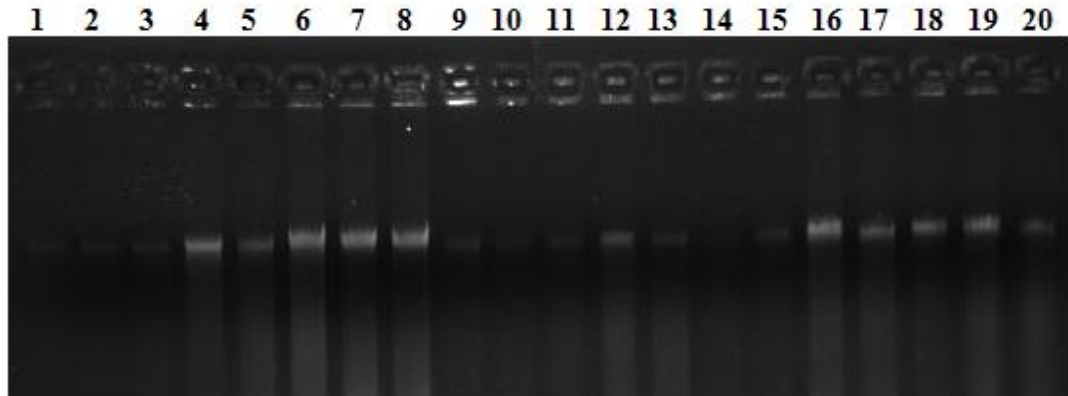
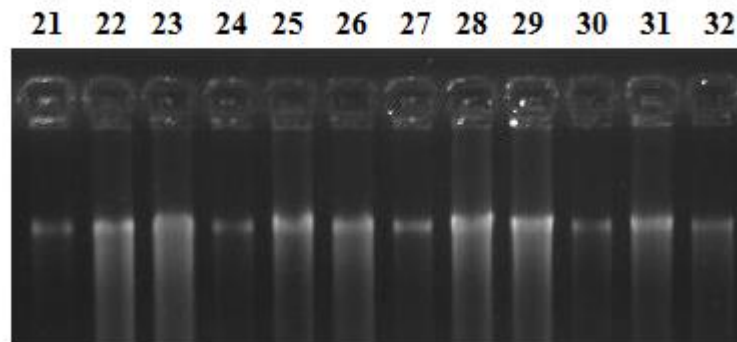


Photo of bacterial viability with the use of LIVE/DEAD® *BacLight* Bacterial Viability Kits and viewed under epifluorescence microscope. Live cells with green fluorescence while dead cells with red fluorescence.

APPENDIX E

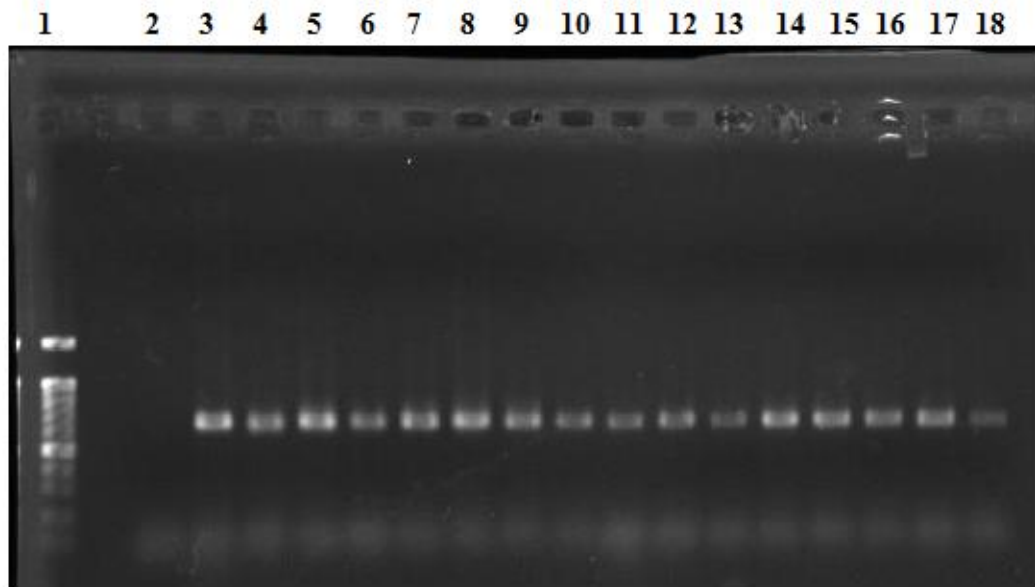


Agarose gel electrophoresis of extracted DNA (1 μ l). Lane 1 to 8 are different samplings from Klang and lane 9 to 20 are different samplings from Port Dickson.



Agarose gel electrophoresis of extracted DNA (1 μ l). Lane 21-23 & 27-29 are different samplings from Klang while lane 24-26 & 30-32 are different samplings from Port Dickson.

APPENDIX F



Agarose gel electrophoresis of amplified 16S rRNA. Lane 1 is 100 bp DNA ladder (iNtRON Biotechnology, Korea), lane 2 is negative control, lane 3 is positive control, lane 4 to 10 are samplings from Klang and lane 11 to 18 are samplings from Port Dickson.

APPENDIX G

Present-absent matrix of the bands on the DGGE gel picture for cluster analysis in Figure 3.5.

OTUs	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
PK(Oct'09)	0	0	0	0	0	0	0	1	0	0	0	0	1	1	1	1	1	1	1	0
PK(Dec'09)	0	0	0	0	0	0	0	1	0	1	1	1	1	1	0	1	1	1	1	0
PK(Jan'10)	0	0	0	0	0	0	0	1	1	1	1	1	0	0	1	1	1	1	1	0
PK(Feb'10)	0	0	0	0	0	0	0	1	1	1	1	1	0	0	0	0	0	0	0	0
PK(Mac'10)	1	0	1	0	0	0	0	1	1	1	1	1	0	0	1	1	0	0	1	0
PK(Apr'10)	0	0	1	0	0	1	0	1	0	1	0	0	0	0	1	0	1	1	1	0
PK(May'10)	0	0	0	0	0	0	0	1	1	1	0	1	0	0	0	1	0	0	0	0
PK(Jun'10)	0	0	0	0	0	0	0	1	0	1	0	1	0	0	0	1	0	0	0	0
PD(Nov'09)	1	0	0	1	0	0	0	0	1	1	1	1	0	0	0	0	0	0	1	0
PD(Dec'09)	1	1	0	1	0	0	1	0	1	1	1	1	0	1	0	0	0	0	1	0
PD(Jan'10)	1	1	0	1	1	0	1	0	1	1	1	1	0	0	0	0	0	0	1	0
PD(Feb'10)	1	1	0	1	1	0	1	0	1	1	1	1	0	0	0	0	0	0	1	0
PD(Mac'10)	1	1	0	0	1	0	1	0	1	1	0	1	0	0	0	0	0	0	1	0
PD(Apr'10)	1	1	0	1	1	0	1	0	1	1	1	1	0	0	0	0	0	0	1	0
PD(May'10)	1	1	0	1	1	0	1	0	0	1	0	1	0	0	0	0	0	0	0	1
PD(Jun'10)	1	1	0	0	1	0	0	0	0	1	0	1	0	0	0	0	0	0	0	1

APPENDIX H

Sequencing results in Table 3.2 were deposited in GenBank under accession codes:

JX193773-JX193793.

Uncultured *Flavobacterium* sp. isolate DGGE gel band D1a 16S ribosomal RNA gene,
partial sequence (JX193773)

CCTATGGGTTGTAAACTGCTTTTATACAGGAAGAAAACTCCGACGTGTCG
GAACTTGACGGTACTGTAAGAATAAGGACCGGCTAACTCCGTGCCAGCAGC
CGCGGTAATACGGAGGGTCCAAGCGTTATCCGGAATCATTGGGTTTAAAGG
GTCCGTAGGCGGTTTTTTAAGTCAGAGGTGAAATCCTGCAGCTCAACTGTAG
AATTGCCTTTGATACTGAAAGACTTGAGTTATTGTGAAGTGGTTAGAATGTG
TGGTGTAGCGGTGAAATGCATAGATATCACACAGAATACCAATTGCGAAGG
CAGATCACTAACAATATACTGACGCTCAGGGACGAAAGCGTGGGTAGCGAA
CAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATGGA

Uncultured cyanobacterium isolate DGGE gel band KD12b 16S ribosomal RNA gene,
partial sequence (JX193774)

GGAGCATACCGCGTGAGGGAAGACGGCCTATGGGTTGTAACCATCTTTCCC
CTTGGAGGAATTTTGACTCTGTACCTGAAGAATAAGCATCGGCTTAACTCCG
ATGCCAGCAGCCGCGGTAACACGGAGGATGCAAGTGTTATCTTGGAATCAC
TGGGCGCAAAGCGTCTGTAACCGGTGAAGCAGGTCAAGTGTGAAATCGTGG
GGCTCAACTTGATAATCGCAGTCTAAACTGTT

Appendix H, continued.

Uncultured beta proteobacterium isolate DGGE gel band K5f 16S ribosomal RNA gene, partial sequence (JX193778)

ATACTCTGTGTGGATGACGGTACCGGCATAAGAAGCACCGGCTAACTACGT
GCCAGCAGCCGCGGTAATACGTAGGGTGCAGCGTTAATCGGAATTACTGG
GCGTAAAGCGTGCGCAGGCGGTTGGGTAAGTTTGATGTGAAATCCCCGGGC
TCAACCTGGGAACTGCATTGAAGACTGTCTGACTAGAGTACGGCAGAGGGG
GGTGGAATTCCACGTGTAGCAGTGAAATGCGTAGAGATGTGGAGGAACACC
GATGGCGAAGGCAGCCCCCTGGGTTCGATACTGACGCTCATGCACGAAAGCG
TGGGGAGCAAACAGGATTAGATACCCTGGTAGTCCACGCCCTAAACGATGT
CAACTAGTTGTTGGTAGGGTTAAACCTATTAGTAACGTAGCTAACGCGTGA
AG

Uncultured bacterium isolate DGGE gel band D1b 16S ribosomal RNA gene, partial sequence (JX193780)

ACCTCCACAAGAAGCCCCGGCCAACCTACCTGCCAGCAGCCGCGGTAATACG
TAGGGGGCGAGCGTTGTCCGATTTATTGGGCGTAAACAGCTCGTAGGGCG
TTCAACAAGTCGGTCGTAAAAGTTCACGGCTCAACCCTGAAATGTTCGATCG
ATACTGTTGTGACTATGATACGGTAGAGGTGAGTGTAATTCCTAGTGTAGCG
GTGAAATGCGTAGATATTCGGAGGAACACCAGTTGCGAAGGCAGCTCACTG
GGCCGCTATCGACGCTGAGGAGCGAAAGCTAGGGCAGCAAACAGGATTAG
ATACCCTGGTAGTCCTAGCTGTAAACGATGGATACTAGACGTAAGAACTGG
ATTGACGGTTTCT

Appendix H, continued.

Uncultured *Marinovum* sp. isolate DGGE gel band D1c 16S ribosomal RNA gene,
partial sequence (JX193782)

CAGTAGCAGGTAAAGAAACCCCGGCTAACTCCGTGCCAGCAGCCGCGGTAA
TACGGAGGGGGTTAGCGTTGTTTCGGAATTACTGGGCGTAAAGCGCGCGTAG
GCGGATTAGAAAGTTGGGGGTGAAATCCCGAGGCTCAACCTCGGAACTGCC
TCCAAAAGTCTAGTCTAGAGTTCGAGAGAGGTGAGTGGAATTCCGAGTGT
AGAGGTGAAATTCGTAGATATTCGGAGGAACACCAGTGGCGAAGGCGGCTC
ACTGGCTCGATACTGACGCTGAGGTGCGAAAGTGTGGGGAGCAAACAGGAT
TAGATACCCTGGTAGTCCACACCGTAAACGATGAATGCCAGTCGTCCGATA
GTATACTATTCGGTGACACACCTAACGGATTAAGCATTCCGCCTGGGGAGT
ACGGTCGCAAGATTA AAACTCA

Uncultured *Roseobacter* sp. isolate DGGE gel band KD11g 16S ribosomal RNA gene,
partial sequence (JX193786)

GATGATAATGACAGTAGCAGGTAAAGAAACCCCGGCTAACTCCGTGCCAGC
AGCCGCGGTAATACGGAGGGGGTTAGCGTTGTTTCGGAATTACTGGGCGTAA
AGCGCGCGTAGGCGGATTAGAAAGTTGGGGGTGAAATCCCGGGGCTCAACC
TCGGAAGTGCCTCCAAAAGTCTAGTCTAGAGTTCGAGAGAGGTGAGTGGA
ATTCCGAGTGTAGAGGTGAAATTCGTAGATATTCGGAGGAACACCAGTGGC
GAAGGCGGCTCACTGGCTCGATACTGACGCTGAGGTGCGAAAGTGTGGGGA
GCAAACAGGATTAGATACCCTGGTAGTCCACACCGTAAACGATGAATGCCA
GTCGTCCGGTAGTATACTATTCGGTGACACACCTAACGGATTAAGCATTCCG
CCTGGGGAGTACGGTCGCAAGATTA AAACTC

Appendix H, continued.

Uncultured delta proteobacterium isolate DGGE gel band K5j 16S ribosomal RNA
gene, partial sequence (JX193788)

AGCCGCGGTAATACGGGGGGTGCAAGCGTTGTTTCGGAATTATTGGGCGTAA
AGCGTGCGTAGGCGGACTTTTAAGTCTGATGTGAAATCCCTCGGCTCAACCG
AGGACGTGCATTGGATACTGGAAGTCTTGAGTCCCGGAGAGGAGAGCGGAA
TTCCTAGTGTAGAGGTGAAATTCGTAGATATTAGGAGGAACACCAGTGGCG
AAAGCGGCTCTCTGGACGGTGACTGACGCTGAGGCACGAAAGCATAGGTAG
CAAACAGGATTAGATACCCTGGTAGTCTATGCCGTAAACGATGGATACTAG
TTGGTGCGGGTATTGACCCCTGCGCTGACGAAGCTAACGCATTAAGTATCCC
GCCTGGGGAGTACGGCCGCAAGGCTAAACT

Uncultured delta proteobacterium isolate DGGE gel band K5k 16S ribosomal RNA
gene, partial sequence (JX193789)

CCGGCTAACTCCGTGCCAGCAGCCGCGGTAATACGGGGGGTGCAAGCGTTG
TTCGGAATTATTGGGCGTAAAGCGTGCGTAGGCGGACCATTAAGTCTGATG
TGAAATCCCTCGGCTCAACCGAGGACGTGCATTGGATACTGGTGGTCTTGA
GTCCCGGAGAGGAGAGCGGAATTCCTAGTGTAGAGGTGAAATTCGTAGATA
TTAGGAGGAACACCAGTGGCGAAAGCGGCTCTCTGGACGGTGACTGACGCT
GAGGTACGAAAGCATAGGTAGCAAACAGGATTAGATACCCTGGTAGTCTAT
GCCGTAAACGATGGACACTAGTTGGTGCGGGTATTGACCCCTGCGCTGACG
AAGCTAACGCATTAAGTGTCCCGCCTGGGGAGTACGGCCGCAAGGCTAAAA
CT

Appendix H, continued.

Uncultured *Comamonas* sp. isolate DGGE gel band K3f 16S ribosomal RNA gene,
partial sequence (JX193790)

CATGACGGTACCGTAAGTAATAAGCACCGGCTAACTACGTGCCAGCAGCCG
CGGTAATACGTAGGGTGCAGCGTTAATCGGAATTACTGGGCGTAAAGCGT
GCCAGGCGGTTATGTAAGACAGGTGTGAAATCCCCGAGCTCAACTTGGGA
ATTGCACTTGTGACTGCATAACTAGAGTGCGGCAGAGGGGGATGGAATTCC
ACGTGTAGCAGTGAAATGCGTAGATATGCGGAGGAACACCGATGGCGAAG
GCAATCCCCTGGGCCTGCACTGACGCTCATGCACGAAAGCGTGGGGAGCAA
ACAGGATTAGATACCCTGGTAGTCCACGCCCTAAACGATGTCAACTGGTTGT
TGGGAATTCACCTTCTCTTTAACCAATCTAACACGTGAAATTGACCGCCTGG
G

Uncultured alpha proteobacterium isolate DGGE gel band K3i 16S ribosomal RNA
gene, partial sequence (JX193791)

CGGCTAACTCCGTGCCAGCAGCCGCGGTAATACGGAGGGGGTTAGCGTTGT
TCGGAATTACTGGGCGTAAAGCGCGCGTAGGCGGATCAGAAAGTTGGGGGT
GAAATCCCGGGGCTCAACCCTGGAAGTGCCTCAAACTCCTGGTCTTGAGT
TCGAGAGAGGTGAGTGGGAATTCGAGTGTAGAGGTGAAATTCGTAGATATT
CGGAGGAACACCAGTGGCGAAGGCGGCTCACTGGCTCGATACTGACGCTGA
GGTGCGAAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGTAGTCCACGC
CGTAAACGATGAATGCCAGACGTCGGGTTGCTTGCAATTCGGTGTACACCT
AACGGATTAAGCATTCCGCCTGGGGAGTACGGTCGCAAGATTA AAACTCAA
AGGAATTGAC

Appendix H, continued.

Uncultured *Pelomonas* sp. isolate DGGE gel band K3j 16S ribosomal RNA gene,
partial sequence (JX193792)

TGACGGTACCGTAAGAATAAGCACCGGCTAACTACGTGCCAGCAGCCGCGG
TAATACGTAGGGTGCAAGCGTTAATCGGAATTACTGGGCGTAAAGCGTGCG
CAGGCGGTGATGTAAGACAGGCGTGAAATCCCCGGGCTCAACCTGGGAATG
GCGCTTGTGACTGCATCGCTGGAGTGCGGCAAAGGGGGATGGAATTCCGCG
TGTAGCAGTGAAATGCGTAGATATGCGGAGGAACACCGATGGCGAAGGCA
ATCCCCTGGGCCTGCACTGACGCTCATGCACGAAAGCGTGGGGAGCAAACA
GGATTAGATACCCTGGTAGTCCACGCCCTAAACGATGTCAACTGGTTGTTGG
GTCTTAGCTGACTCAGTAACGAAGCTAACGCGTGAAGTTGACCGCCTGGGG
AGTACGGCCGCA

Uncultured *Donghicola* sp. isolate DGGE gel band K4h 16S ribosomal RNA gene,
partial sequence (JX193793)

GCTAACTCCGTGCCAGCAGCCGCGGTAATACGGAGGGGGTTAGCGTTGTTC
GGAATTACTGGGCGTAAAGCGCGCGTAGGCGGACCAGAAAGTTGGGGGTG
AAATCCCCGGGGCTCAACCCCGGAACGGCCTCCAAAACCTCCTGGTCTTGAGT
TCGAGAGAGGTGAGTGGAATTCCGAGTGTAGAGGTGAAATTCGTAGATATT
CGGAGGAACACCAGTGGCGAAGGCGACTCACTGGCTCGATACTGACGCTGA
GGTGCGAAAGTGTGGGGAGCAAACAGGATTAGATACCCTGGTAGTCCACAC
CGTAAACGATGAATGCCAGTCGTCGGCGCGCATGCGTGTCCGGTGACACACC
TAACGGATTAAGCATTCCGCCTGGGGAGTACGGTCGCAAGATTA AAACTCA
AAG
