

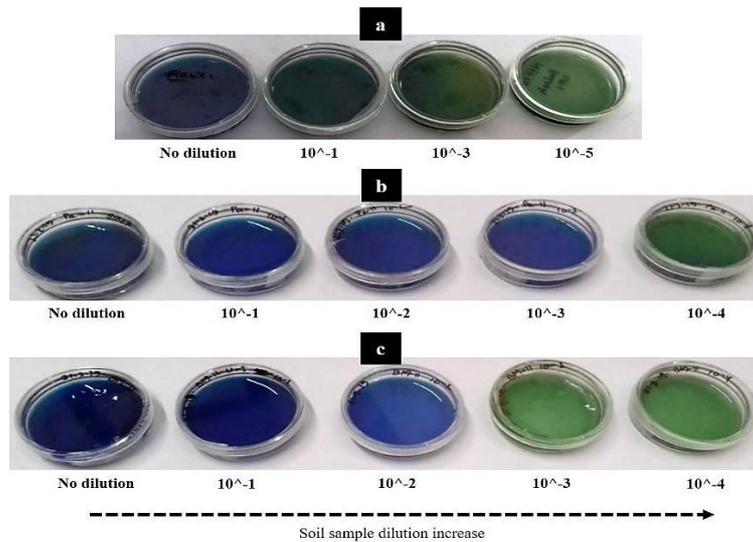
Screening and isolation of potential nitrogen-fixing *Enterobacter* sp. GG1 from mangrove soil with its accelerated impact on green chili plant (*Capsicum frutescens* L.) growth amelioration

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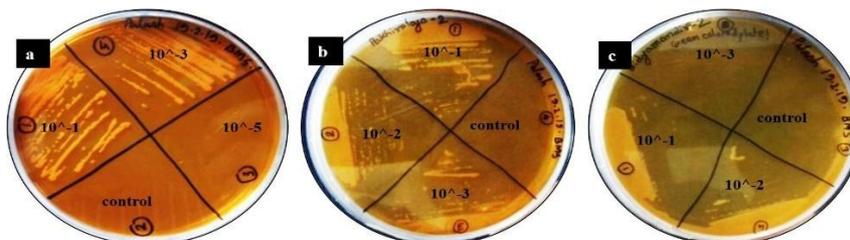
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doi: <https://doi.org/10.7324/JABB.2023.139674>

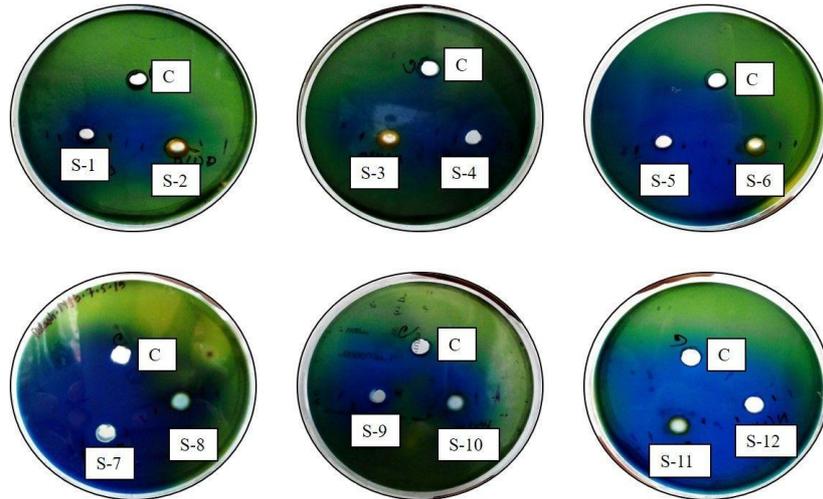
Supplementary Figures:



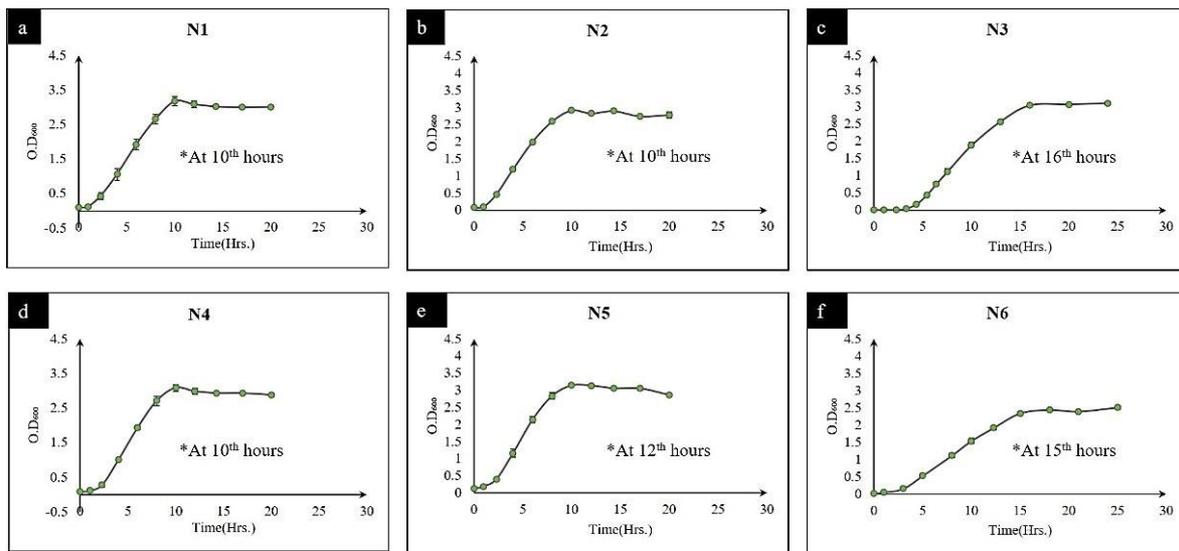
Supplementary Figure S1: Isolation of N-fixing bacteria in Nfb semi-solid medium (a: Dobanki sample; b: Pakhiralay sample; c: Mathurakhanda sample)



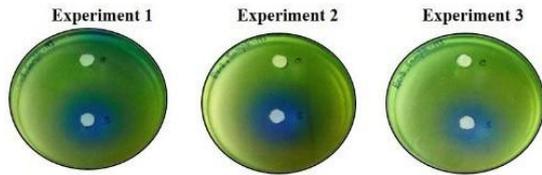
Supplementary Figure S2: Streak in BMS medium for N-fixing bacterial single colony isolation (a: Dobanki sample; b: Pakhiralay sample; c: Mathurakhanda sample)



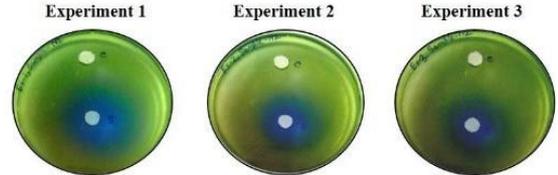
Supplementary Figure S3: Preliminary study of primary screening by well diffusion method in Nfb agar plate (C=control; S=sample).



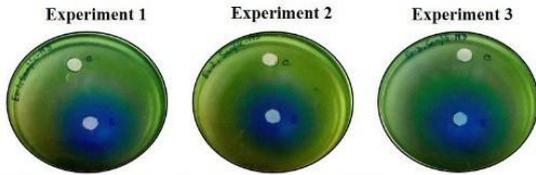
Supplementary Figure S4. Exponential phase study of nitrogen-fixing bacteria in Nfb media.



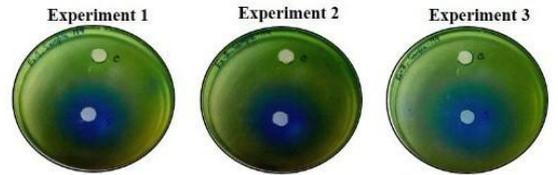
a: N1 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)



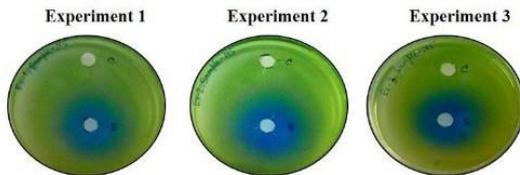
b: N2 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)



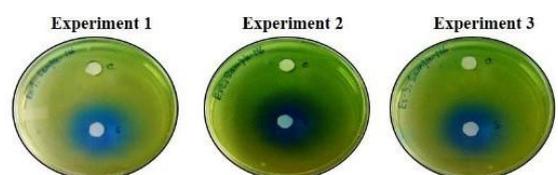
c: N3 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)



d: N4 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)

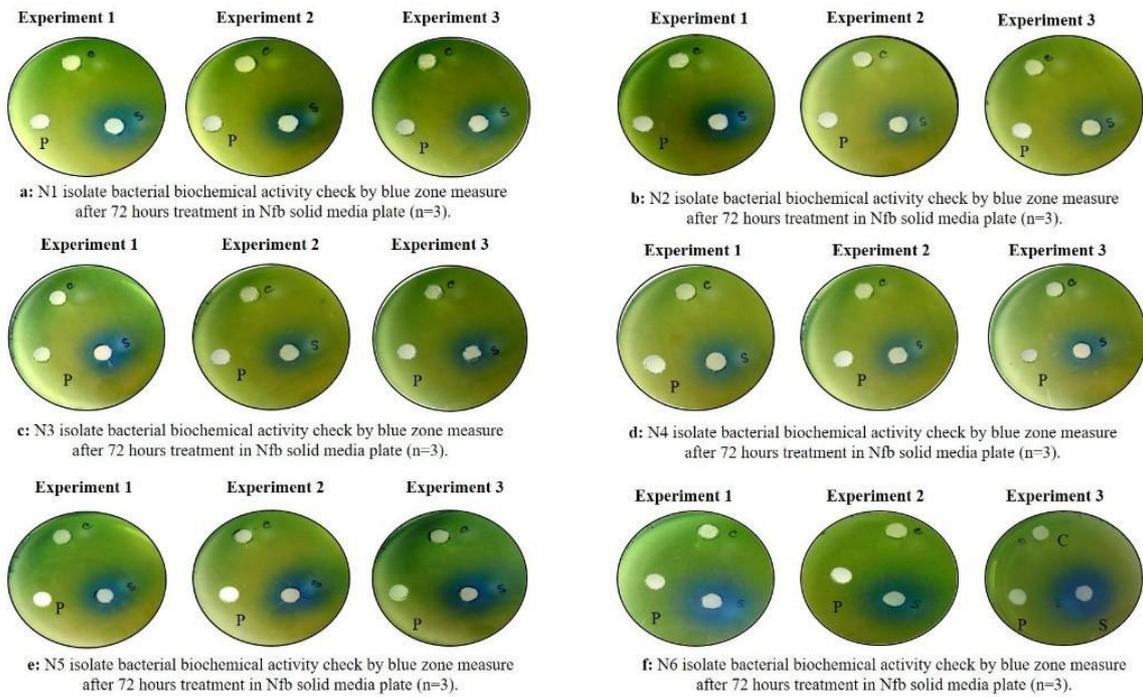


e: N5 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)

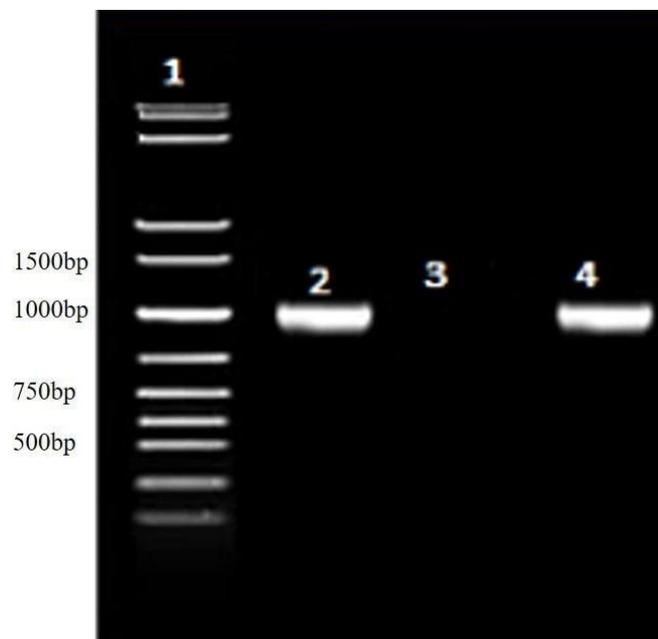


f: N6 isolate bacterial N_2 -fixation ability check by blue zone measure after 72 hours treatment in Nfb solid media plate (n=3)

Supplementary Figure S5: Exponential phase study of nitrogen-fixing bacteria in Nfb media (C= control; S= isolated culture).



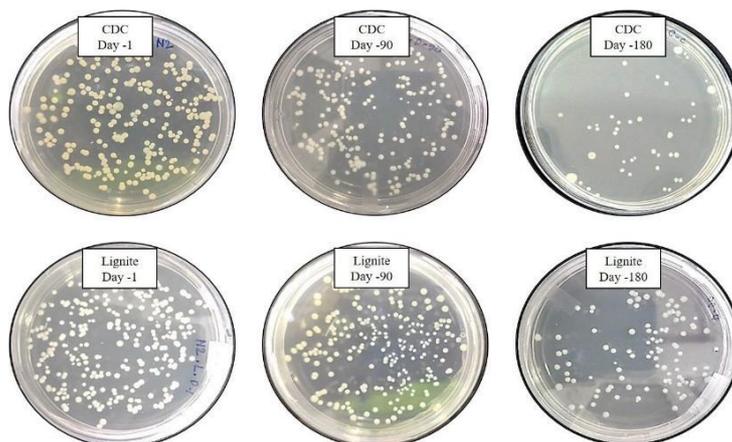
Supplementary Figure S6: Determine the extracellular or intracellular response for N-fixation in Nfb media; when, C= PBS buffer as a control, P= crude extract of N-fixing bacterial isolate (for intracellular response investigation), S= N-fixing bacterial alive cell (for extracellular response investigation).



Supplementary Figure S7: Shows a single 1Kb PCR product of 16S rRNA gene on 2% agarose gel (Lane 1: 1KB DNA ladder, Lane 2: 16S amplicon (Positive Control: *E. coli*), Lane 3: Negative Control, Lane 4: Sample).

Description	Max Score	Total Score	Query Cover	E value	Per. Ident
Enterobacter sp. FF16 16S ribosomal RNA gene, partial sequence	2346	2346	100%	0.0	98.93%
Enterobacter cloacae strain ACD1 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter mori strain AS5 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter ludwigii strain AA1 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter tabaci strain NAC74 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter sp. strain LSB19 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter sp. strain LSB10 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter roggenkampii CCI9 gene for 16S ribosomal RNA, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter asburiae strain TY175-13 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%
Enterobacter ludwigii strain TY175-12 16S ribosomal RNA gene, partial sequence	2340	2340	100%	0.0	98.86%

Supplementary Figure S8: 16S rRNA sequence of N4 bacterial isolates was generated BLAST analysis to compare with the sequences in the NCBI GenBank database. The N4 isolate has been higher similar to *Enterobacter* sp. bacterial strain.



Supplementary Figure S9: Best plat show of viability study at 180 days of *Enterobacter* sp. GG1 in carriers (CDC and lignite).

Supplementary Data file SD1

Enterobacter sp. GG1_16s rRNA gene => Enterobacter sp. GG1

TGTGTACAAGCCCCGGGAACGTATTCACCGTAGCATTCTGATCTACGATTACTAGCGATTCCGACT
TCATGGAGTCGAGTTGTCAGACTCCAATCCGGACTACGACGCATTTTATGAGGTCCGCTTGCTCTCG
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ACGCGTTAGCTCCGGAAGCCACGCCTCAGGGGCACAACCTCCAAGTCGACATCGTTTACGGCGTG
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TCCGTATTACCGCGGTGCTGGCACGGAGTTAGCCGGTGTCTTCTGCGGGTAACGTCATTCGCT
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AAGTCCCCCTCTTTGGTCTTGCAGCTTATGCGGTATTAGCTACCGTTCCAGTAGTTATCCCCCT CC
ATCAGGCAGTTTCCAGACATTACTACCAATGCGCCGCTCGCCGGCAAAGTAG

Azotobacter chroococcum YSC6 => AB696769.1:798-1494 Azotobacter chroococcum gene for 16S ribosomal RNA, partial sequence, strain: NTUIOB YSC6

ACGCCGTAACGATGTCGACTAGCCGTTGGGCTCCTTGAGAGCTTGGTGGCGCAGCTAACGCATTA
AGTCGACCGCCTGGGGAGTACGGCCGCAAGGTTAAAACCTCAAATGAATTGACGGGGGCCCGACA
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CTCGGGTGGGCACTTAAGGAGACTGCCGGTGACAAACCGGAGGAAGGTGGGGATGACGTCGAAGT
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CGCGAGGTGGAGCTAATCCAGAAAACCGATCGTAGTCCGGATCGCAGTCTGCAACTCGACTGCG
TGAAGTCGGAATCGCTAGTAATCGCGAATCAGAATGTCGCGGTGAATACGTTCCCGGGCCTTGTAC
ACACCGCCGTCACACCATGGGAGTGGGTTGCTCCAGAAGTAGCTAGTCTAACCTTCGGGGGGAC
GGTTACCACGGAGTGATTATGACTGGGGTGAAGTCGTAACAA

Azospirillum lipoferum B7=> AY685925.1:1-281 Azospirillum lipoferum clone B7 16S-23S intergenic spacer, complete sequence

CTAAGGAAGCCGACCCTGGTGGGTCCGGCACCTTCAAGTCCAGATGGCGCATCTCTGCCGCCGCCG
GCGCATCCCTTCTCGACGATCCGGAACACCCGCTGACAGTGCATCCGCACTGTCTCGGCCGGATTG
ACGAGAAGCGCTTGGCGTTTGAATTTTCAAACGCGGGATCTTCAAATCGTGAATAAAGTCGA
GTTTTAAGTGACCGAGGATGCATCTCATGAGCGTCCACGAGGCAGGAGCGGTTTGGCCCTGTGCG
TGACGTTCCGGAATAGAG

Beijerinckia sp. strain 37L => MG904942.1:1-1317 Beijerinckia sp. strain 37L 16S ribosomal RNA gene, partial sequence

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GGTAAAGGCCTACCAAGGCGACGATCCATAGCTGGTCTGAGAGGATGATCAGCCACATTTGGGACT
GAGACACGGCCAAACTCCTACGGGAGGCAGCAGTGGGGAATATTGGACAATGGGGCGCAGCCTG
ATCCAGCCATGCCCGTGAGTGTGATGAAGGCCTTAGGGTTGTAAAGCTTTTACCAGGAGAAGATA
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CATGAAGTTGGAATCGCTAGTAATCGCAGCTTGGGTACGCGACTGGGGTGAATCTACAAGAGAGA
GGCCCCCATAAAA

Burkholderia sp. strain OKR4-1 => OP872624.1:1-1502 Burkholderia sp. strain OKR4-1 16S ribosomal RNA gene, partial sequence

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ACATGTCTGTAGTGGGGGATAGCCCGGCGAAGCCGGATTAATACCGCATAACGATCTACGGATG
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GCACTCTGCAACTCGAGTGCATGAAGCTGGAATCGCTAGTAATCGCGGATCAGCATGCGCGGGGG
GAGAAACCTTCCCCGGGTTCTTGTACACACCCGCCGTCACACCATGGGAAGTGGGTTTTACCAGAA
GTGGCTAGTCTAACCGCAAGGAGGACGGTACCACGGTAGGATTCTGACTGGGTGAGCAAAAA

Clostridium sp. M98449 => M98449.1:1-137 Clostridium sp. 16S ribosomal RNA (16S rRNA) gene, partial sequence

GAGATTAGGAAGAACCCAGTGGCGAAGGCGGCTTTCTGGACTGTAACCTGACGCTGAGGCCCGAA
AGCGTGGGGAGCAAACAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATGAGTACTAGGTG TAGGAGG

Enterobacter cloacae strain OsEp_A&N_15A7=> MT367840.1:12-1397 Enterobacter cloacae strain OsEp_A&N_15A7 16S ribosomal RNA gene, partial sequence

GAACGGTAGCACAGAGAGCTTGTCTCGGGTGACGAGTGGCGGACGGGTGAGTAATGTCTGGGAA
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Gluconacetobacter sp. 0w26 => OK641906.1:1-579 Gluconacetobacter sp. strain 0w26 16S ribosomal RNA gene, partial sequence GTAACGCGTAGGAATCTATCTTTGGGTGGGGGATAACTGTGGGAAACTACAGCTAATACCGCATG AAACCTCAGGGTCAAAGCAGCGATGCGCCGAAGGAGGAGCCTGCGTACGATTAGCTAGTTGGTGG GGTAAAGCCCTACCAAGGCTACGATCGTTAGCTGGTCTGAGAGGATGATCAGCCACATTGGGACT GAGACACGGCCAAACTCTACGGGAGGCAGCAGTGGGGAATATTGGACAATGGGGCAAAGCCT GATCCAGCAATGCCGCGTGTGTGAAGAAGTCTTCGGATTGTAAAGCACTTTCGACGGGGACGAT GATGACGGTACCCGTAGAAGAAGCCCCGGCTAACTTCGTGCCAGCAGCCGCGGTAATACGAAGGG GGCTAGCGTTGCTCGAATGACTGGGCGTAAAGGGCGCGTAGGCGGATTGTACAGTCAGATGTGA AATTCCCGGGCTAACCTGGGGGCTGCATTTGATACGTATAGTCTAGAGTGAGGAAGAGGGTTGTG GAATCCCAGTGTAGAGGTGAAATTCGTAGATATTGGGAAGAACACCGGTGGCGAAGGC

Herbaspirillum sp. Ah5_2.5 => GQ181144.1:13-488 Herbaspirillum sp. enrichment culture clone Ah5_2.5 16S ribosomal RNA gene, partial sequence TAGGAGCTTGCTCCTGATGGCGAGTGGCGAACGGGTGAGTAATATATCGGAACGTGCCCTAGAGT GGGGGATAACTAGTTCGAAAGATTAGCTAATACCGCATACGATCTAAGGATGAAAGTGGGGGATCG CAAGACCTCATGCTCCTGGAGCGGCCGATATCTGATTAGCTAGTTGGTGGGGTAAAAGCCTACCAA GCGACGATCAGTAGCTGGTCTGAGAGGACGACCAGCCACACTGGGACTGAGACACGGCCACAGAC TCCTACGGGAGGCGACTGGGGAATTTGGACAATGGGGGAACCCCTGATCCAGCAATGCCCGG TGAGTGAAGAAGGCTTCGGGTTGTAAGCTTTTGTTCAGGGAAGAAACGGTTCCAGCTAATATC TGGAGCTAATGACGGTACCTGAAGAATAAGCACCGGCTAACTACGTGCCAGCAGCCGCGGTAATA CGTAGGGTGAAGCGTTAA

Ideonella sp. URA-8=> LC505027.1:1-1471 Ideonella sp. URA-8 gene for 16S ribosomal RNA, partial sequence TGGCTCAGATTGAACGCTGGCGGCATGCCTTACACATGCAAGTCGAACGGTAACGCGGGCAACC TGGCGACGAGTGGCGAACGGGTGAGTAATGCATCGGAACGTGCCAGTAGTGGGGGATAGCCCGG CGAAAGCCGGATTAATACCGCATAACGACCTGAGGGTAAAGCGGGGGATCGCAAGACCTCGCGCT ATTGGAGCGGCCGATGTCAGATTAGGTAGTTGGTGGGGTAAAGGCCTACCAAGCCGACGATCTGT AGCTGGTCTGAGAGGACGACCAGCCACACTGGGACTGAGACACGGCCCAGACTCCTACGGGAGGC AGCAGTGGGGAATTTGGACAATGGGCGCAAGCCTGATCCAGCCATGCCGCGTGCGGGAAGAAGG CCTTCGGGTTGTA AACCCGCTTTTGTTCAGGGAAGAAATCCTTTGGGCTAATACCCCGGGGGGATGAC GGTAACTGAAGAATAAGCACCGGCTAACTACGTGCCAGCAGCCGCGGTAATACGTAGGGTGCAG CGTTAATCGGAATTA CTGGGCGTAAAGCGTGCAGCGGCTTTTGTAAAGACAGAGGTTGAAATCCC CGGGCTCAACCTGGGAACTGCCTTTGTGACTGCAAGGCTTGAGTGCGGCAGAGGGGGATGGAATT CCGCGTGTAGCAGTGAATGCGTAGATATGCGGAGGAACACCGATGGCGAAGGCAATCCCCTGGG CCTGCACTGACGCTCATGCACGAAAGCGTGGGGAGCAAACAGGATTAGATAACCTGGTAGTCCAC GCCCTAAACGATGTCAACTGGTTGTTGGGAAGGTTCTTCTCAGTAACGTAGCTAACCGGTGAAGT TGACCGCTGGGAGTACGGCCGCAAGGTTGAAACTCAAAGGAATTGACGGGGACCCGCACAAGC GGTGGATGATGTGGTTAATTCGATGCAACGCGAAAAACCTTACCTACCCTTGACATGCCTGGAAT CCTGCAGAGATGTGGGAGTGTCTGAAAGAGAGCCAGGACACAGGTGCTGCATGGCCGTCGTCAGC TCGTGTCTGAGATGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCCTGTCATTAGTTGCTACGAAA GGGCACTCTAATGAGACTGCCGGTGACAAACCGGAGGAAGGTGGGGATGACGTCAGGTCCTCATG GCCCTTATGGGTAGGGCTACACACGTCATAACAATGGCCGGTACAGAGGGCTGCCAACCCGCGAGG GGGAGCCAATCCCAGAAAACCGGTTCGTAGTCCGGATCGCAGTCTGCAACTCGACTGCGTGAAGTC GGAATCGCTAGTAATCGCGGATCAGCTTGCCGCGGTGAATACGTTCCCGGGTCTTGTACACACCGC CCGTACACCATGGGAGCGGGTCTGCCAGAAGTAGTTAGCCTAACCGCAAGGAGGGCGATTACC ACGGCAGGGTTTCGTGACTGGGGTGAAGTCGTAACAA

Klebsiella sp. SXW12 => OP648246.1:1-1418 Klebsiella sp. strain SXW12 16S ribosomal RNA gene, partial sequence AGCTACACATGCAGTCGAGCGGTAGCACAGAGAGCTTGCTCTCGGGTGACGAGCGGCGGACGGGT GAGTAATGTCTGGGAAACTGCCTGATGGAGGGGGATAACTACTGGAAACGGTAGCTAATACCGCA TAACGTCGCAAGACCAAAGTGGGGGACCTTCGGGCCCTATGCCATCAGATGTGCCAGATGGGAT TAGCTGGTAGGTGGGGTAAACGGCTCACCTAGGCGACGATCCCTAGCTGGTCTGAGAGGATGACCA GCCACACTGGAACCTGAGACAGGTCAGACTCTACGGGAGGCAGCAGTGGGGAAATATTGCACAA TGGGCGCAAGCCTGATGCAGCCATGCCGCGTGTGTGAAGAAGGCCTTCGGGTTGTAAGACTTC TCAGCGGGGAGGAAGGCGGTGAGGTTAATAACCTCATCGATTGACGTTACCCGCAAGAAGAAGCACCG GCTAACTCCGTGCCAGCAGCCGCGTAATACGGAGGGTGAAGCGTTAATCGGAATTA CTGGGCG TAAAGCGCACGCAGGCGGTCTGTCAAGTCGGATGTGAAATCCCCGGGCTCAACCTGGGAACTGCA TTCGAAACTGGCAGGCTAGAGTCTTGTAGAGGGGGTAGAATTCCAGGTGTAGCGGTGAAATGCG TAGAGATCTGGAGGAATACCGGTGGCGAAGGCGGCCCCCTGGACAAAGACTGACGCTCAGGTGCG AAAGCGTGGGGAGCAAACAGGATTAGATAACCTGGTAGTCCACGCTGTAAACGATGTCGATTTGG AGGTTGTGCCCTTGAGGCGTGGCTTCCGGAGCTAACCGGTTAAATCGACCGCCTGGGGAGTACGG CCGCAAGGTTAAAACCTAAATGAATTGACGGGGGCCCCACAAGCGGTGGAGCATGTGGTTTAAAT TCGCAACGCAAGCAAGAACCTTACCTGGTCTTGACATCCACAGAACTTCCAGAGATGGATTGGTG CCTTCGGGAACTGTGAGACAGGTGCTGCATGGCTGTGCTGACGTCGTTGTGAAATGTTGGGTTA AGTCCCAGCAACGAGCGCAACCCCTTATCCTTTGTTGCCAGCGGTTAGGCCGGAACTCAAAGGAGA CTGCCAGTGATAA ACTGGAGGAAGGTGGGGATGACGTCAGTCAAGTCATCATGGCCCTTACGACCAGGG CTACACACGTGCTACAATGGCATATACAAGAGAAGCGACCTCGCGAGAGCAAGCGGACCTCATA AAGTATGTCGTAGTCCGGATTGGAGTCTGCAACTCGACTCCATGAAGTCGGAATCGCTAGTAATCG TAGATCAGAATGCTACGGTGAATACGTTCCCGGGCCTTGTACACACCGCCGTCACACCATGGGAG

TGGGTTGCAAAAAGAAGTAGGTAGCTTAACCTTCGGGAGGGCGCTACCA

Methanosarcina sp. T40UC2 => MG008506.1:1-1135 Methanosarcina sp. strain T40UC2 16S ribosomal RNA gene, partial sequence

TGCTGGAATGCTTTATGCGTCAAATGGATTTCGTCTGCCCAAGGATGGGTCTGCGGCCTATCAGGTA
GTAGTGGGTGTAATGTACCTACTAGCCGACAACGGGTACGGGTTGTGAGAGCAAGAGCCCGGAGA
TGGATTCTGAGACATGAATCCAGGCCCTACGGGGCGCAGCAGGCCGCAAAAACTTTACAATGCGGG
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AACGGGTGGAGCCTGCGGTTTAAATTGGACTCAACGCCGGACAACCTACCGGGGGCGCAGCAATA
TGTAGCCAAAGTGAAGACTTTGCCTGAATCGTAGAGGAGGTGCATGGCCGTCGCCACTTCGT
ACTGTGAAGCATCTGTTAAGTCAAGCAACGAGCGAGACCCGTGCCACTGTTACCAGCATATTCT
CCGGAATGATGGGTACTCTGTGGGGACCGCTATGTTAAATAGGAGGAAGGTGCGGGCCACGGTA
GGTCAGTATGCCCCGAATCTCCCGGGCTACACGCGGGCTACAATGGATGGGACAATGGGTCCCTC
CCCCGAAAGGGCTGGTAATCTCACAACCCATTTCGTAGTTCGGATCGAGGGCTGTAACCTCGCCCT
CGTGAAGCTGGAATCCGTAGTAATCGC

Pseudomonas sp. strain S'23 => MW578905.1:26-1387 Pseudomonas sp. strain S'23 16S ribosomal RNA gene, partial sequence

GTCGAGCGGATGAGAGGAGCTTGCTCCTTGATTTAGCGGCGGACGGGTGAGTAATGCCTAGGAAT
CTGCCTGGTAGTGGGGGATAACGTTCCGAAAGGAACGCTAATACCGCGTACGTCTACGGGAGAA
AGCAGGGGACCTTCGGGCCTTGCCTATCAGATGAGCCTAGGTTCGGATTAGCTAGTTGGTGAGGT
AATGGCTACCAAGGCGACGATCCGTAACCTGGTCTGAGAGGATGATCAGTCACACTGGAAGTGA
ACACGGTCCAGACTCCTACGGGAGGCAGCAGTGGGGAATATTGGACAATGGGCGAAAGCCTGATC
CAGCAATGCCCGTGTGTGAAGAAGGTCTTCGGATTGTAAGCACTTTAAGTTGGGAGGAAGGGC
AGTAAGTTAATACCTTGGCTGTTTGGACGTACCGGACAGAATAAGCACCGGCTAACCCTCGCCAGC
AGCCCGGTAATACGAAGGGTGAAGCGTTAATCGGAATTACTGGGCGTAAAGCGCGCGTAGGTG
GTTTCGTTAAGTTGGATGTGAAAGCCCCGGGCTCAACCTGGGAACTGCATCCAAAACCTGGCGAGCT
AGAGTACGGTAGAGGGTGGTGGAATTTCTGTGTAGCGGTGAAATGCGTAGATATAGGAAGGAAC
ACCAGTGGCGAAGGCGACCACCTGGACTGATACTGACACTGAGGTGCGAAAGCGTGGGGAGCAA
ACAGGATTAGATACCTGGTAGTCCACGCCGTAACGATGTCAACTAGCCGTTGGAATCCTTGAGA
TTTTAGTGGCGCAGCTAACGCATTAAGTTGACCGCTGGGAGTACGGCCGCAAGGTTAAAACCTC
AAATGAATTAGCGGGGCGCACAAGCGGTGAGCATGTGGTTTAAATTCGAAGCAACGCGAAGCA
ACCTTACCTGGCCTTGACATGCTGAGAACTTTCCAGAGATGGATTGGTGCCTTCGGGAACTCAGAC
ACAGGTGCTGCATGGCTGTCGTCAGCTCGTGTGCTGAGATGTTGGGTTAAGTCCCCTAACGAGCGC
AACCTTGTCTTATGTTACCAGCACCTCGGGTGGGCACTCTAAGGAGACTGCCGGTGACAAACCGG
AGGAAGGTGGGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGGGCTACACACGTGCTACAAT
GGTCCGTACAAAGGGTTGCCAAGCCGCGAGGTGGAGCTAATCCATAAAAACCGATCGTAGTCCGG
ATCGCAGTCTGCAACTCGACTGCGTGAAGTCGGAATCGCTAGTAATCGTGAATCAGAATGTCACG
GTGAATACGTTCCCGGGCCTTGTACACACCCGCCGTCACACCATGGGAGTGGGTTGCT

Paenibacillus chibensis SK4-3.2 => MN421117.1:179-1267 Paenibacillus chibensis strain SK4-3.2 16S ribosomal RNA gene, partial sequence

CGGAGCAACGCCGCGTGAGTGATGAAGGTTTTCCGGATCGTAAAGCTCTGTTGCCAGGGAAGAACG
TCCGGTAGAGTAACTGCTACCGGAGTGACGGTACCTGAGAAGAAAGCCCCGGCTAATTACGTGCC
AGCAGCCGCGGTAATACGTAGGGGGCAAGCGTTGTCCGGAATTATTGGGCGTAAAGCGCGCGCAG
GCGGTCACTTAAGTCTGGTGTTTAAGGCCAAGGCTCAACCTTGGTTGCACTGGAAACTGGGTGAC
TTGAGTGCAGAAGAGGAGAGTGAATTCACGTGTAGCGGTGAAATGCGTAGATATGTGGAGGAA
CACCAGTGGCGAAGGCGACTCTCTGGGCTGTAACCTGACGCTGAGGCGCGAAAGCGTGGGGAGCAA
ACAGGATTAGATACCTGGTAGTCCACGCCGTAACGATGAATGCTAGGTGTTAGGGGTTTCGATA
CCCTTGGTGGCGAAGTTAACACATTAAGCATTCCGCCTGGGGAGTACGGTCGCAAGACTGAAACTC
AAAGGAATTGACGGGGACCCGCACAAGCAGTGGAGTATGTGGTTTAAATTCGAAGCAACGCGAAGA
ACCTTACCAAGTCTTGACATCCCTCTGAATCCCTAGAGATAGAGGCGGCCCTTCGGGACAGAGGTG
ACAGGTGGTGCATGGTTGTCGTCAGCTCGTGTGCTGAGATGTTGGGTTAAGTCCCCTAACGAGCGC
AACCTTGTATTTAGTTGCCAGACTTCCGGTGGGCACTCTAGAATGACTGCCGGTGACAAACCCG
AGGAAGGCGGGGATGACGTCAAATCATCATCCCTTATGACTTGGGCTACACACTACAAT
GGCCAGTACAACGGGAAGCGAAGTCGCGAGATGGAGCCAATCCTATCAAAGCTGGTCTCAGTTCG
GATTGCAGGTGCAACCCGCTGCATGAAGTCGGAATTGCTAGTAATCGCGGATCAGCATGCCGC
GGTGAATACGTTCCCGGGTCTTGTACACACCCGCCGTCACACCACGAGAGTTTACAACACCCGAAG
TCGGTGGGTAACCCGCAAGGGAGCCAGCCGCCGAAGGTGGG

Supplementary Data file SD2: Multiple sequence alignment of N-fixing free living bacterial based on 16s rRNA gene sequences



Consensus CA0AG0ACATGGACTGAGA0ACGGC0AA0T0C -TACGGGAGGCAGCAGTGGGG-

Azospirillum lipoferum B7 21
 B akh odeni.sp.OKR4+a 367
Enterobacter *doeae* OsEp_AN_15A7 297
Glucosacetobacter sp. Dw26 312
Ideonella sp. URA-8 335
Klebsiella sp. SXW12 312
Pseudomonas sp. S23 298

A T T C A T A C T T G A C C C T T C

Durkholderia sp. OKR4-1 430
Glucosacetobacter sp. Dw26 297
Ideonella sp. URA-8 398
Methanosarcina sp. T40UC2 558
Pseudomonas sp. S23 30
Paenibacillus chibensis SK4-3.2

A A A C T T T A C A A T G C G G A A A C C G T A A C G G A C A C C G A 0 T G T C A - - - - - 0 C A T C A T A T G C T H G C T G C C A G 0 T

Enterobacter sp. GG1 450
B akh rodens sp. OKR4-a 510
Enterobacter *doeae* OsEp_AN_15A7 440
Glucosacetobacter sp. Dw26 352
Ideonella sp. URA-8 470
Klebsiella sp. SXW12 438
Paenibacillus chibensis SK4-3.2 116

C A C T C C C C C T A T C C A T T C A T A T C T A A C

Enterobacter sp. GG1 520
Azospirillum lipoferum B7 87
B akh rodens sp. OKR4+a 509
Enterobacter *doeae* OsEp_AN_15A7 537
Glucosacetobacter sp. Dw26 478
Herbaspirillum sp. RhS_2.5 557
Ideonella sp. URA-8 525
Klebsiella sp. SXW12 510
Pseudomonas sp. S23

Consensus
 A...CGGCOTGGGGAGFA0G5 0G0AA...GTTAAA0F0AAA...GAATW6ACGGGG...004G0-A0AAGGG

Azotobacter chroococcum T5Cb
 GACCGCCTGGCTGCCTTTACGCCAGTAATTCGGATTAAACGCTAGCACCCCTCGTATTACCGCGG--CTGCTGGCACGG 12s
 AGTCGACCGCCTGGGGAGTACGCCCGCAA-----GGTTAAACTCAAAA-----TGAATTGACGGGGGCCCGC-ACAAAGCG ia7

Beijerinckia sp. 37L
 AACATTCCGCTGGGGAGTACGGTCGCAA-----GATTAAACTCAAAA-----GGAATTGACGGGGGCCCGC-ACAAAGCG a49

Burkholderia sp. OKR4-1
 AGTTGA CCGCCTGGGGAGTACGGTCGCAA-----GATTAAACTCAAAA-----GGAATTGACGGGGGCCCGC-ACAAAGCG sfs

Enterobacter doecae OsEp_AN_15A7
 AATCGACCGCCTGGGGAGTACGCCCGCAA-----GGTTAAACTCAAAA-----TGAATTGACGGGGGCCCGC-ACAAAGCG 876

Ideonella sp. URA-8
 AGTTGA CCGCCTGGGGAGTACGCCCGCAA-----GGTTAAACTCAAAA-----GGAATTGACGGGGGCCCGC-ACAAAGCG 9y2

Klebsiella sp. SXW12
 AATCGACCGCCTGGGGAGTACGCCCGCAA-----GGTTAAACTCAAAA-----TGAATTGACGGGGGCCCGC-ACAAAGCG 892

Methanosarcina sp. T4OUC2
 AGCGAGCCACTGGGAAGTACGGCCGCAA-----GGCTGAAACTTAAA-----GGAATTGGCGGGGGAGCACAAACAGCG 721

Pseudomonas sp. S23
 AGTTGA CCGCCTGGGGAGTACGCCCGCAA-----GGTTAAACTCAAAA-----TGAATTGACGGGGGCCCGC-ACAAAGCG 8y2



Enterobacter sp. GG1
 eGzPAGCCGGTGCFSPYI CTGGGGI AACGvCé *YcGcvGAs GFIAT YACCITSCAa Ca CCYrc-----YC 952

Azotobacter chroococcum Y5Cb
 GYGGGGcEY GYGG L YAMY CGAA CAACGCCGA-----sAAeCTYAeCzsccY-----?GA 10a

Beijerinckia sp. 37L
 GTGGAGCATGTGGTTAATTCGAAAGCAACGCCGA-----GAACCTTACCAGCTCT-----TGA 902

Burkholderia sp. OKR4-1
 GTGGATGATGTGGATTAATTCGATGCAACGCCGA-----AAACCTTACCCTACCT-----TGA 888

Pseudomonas sp. S23
 GTGGATGATGTGGTTAATTCGATGCAACGCCGA-----AAACCTTACCCTACCT-----TGA 928

Paenibacillus chibensis SK4-3.2
 GTGGAGTATGTGGTTAATTCGAAAGCAACGCCGA-----GAACCTTACCAGTCT-----TGA 603



Enterobacter sp. GG1
 CCC-GCTGAAAGT--ACTTTACAACCCGAAGCCCTTCTC--AT-----ACACGGGCATGGGTGCATCAGGC 1ajs

As e bacter chiooi horn
 CAT-CCCTGCGAAC--CTTCCAGAGATGAGAGGCTGCTTC--GGAACTGTGAGACAGGTGCTGCATGGCTGTCTGCAGC 2aj

As @ iFlum lipoferum W
 CAT-TCGGGATA TGGGCATTGGAGACGATGCTCTCAGTTAGCTGGCCCAAGACAGGTGCTGCATGGCTGTCTGCAGC i?q

Burkholderia sp. OKR4-1
 CAT-GGTCGGAAT--CCGCTGAGAGGTGGAGTGTCTCGAAAAGAGAACCGCGCACAGTGTCTGCATGGCTGTCTGCAGC loss

Ideonella sp. URA-8
 CAT-GCCTGGAAT--CCTGCAGAGATGTTGGAGTGTCTCGAAAAGAGGCCAGGACACAGGTGCTGCATGGCTGTCTGCAGC 104j

Klebsiella sp. SXW12
 CAT-CCACAGAAC--TTTCCAGAGATGGATTGTGCTTC--GGAACTGTGAGACAGGTGCTGCATGGCTGTCTGCAGC 102q

Pseudomonas sp. S23
 CAT-GCTGAGAAC--TTTCCAGAGATGGATTGTGCTTC--GGAACTCAGACACAGGTGCTGCATGGCTGTCTGCAGC i0os

Paenibacillus chibensis SK4-3.2
 CATCCCTCTGAAT--CCTCTAGAGATAGAGGCG--GCCTTC--GGACAGAGGTGACAGGTGCTGCATGGTGTCTGCAGC 69u



Consensus
 TGGTG...TGTGAATGTT*...GGGFFAAGT000G0A0GAG0 G0A000TTG:G TF:GFT'00AG

Enterobacter sp. GG1
 TTCCGCCCATTTGTCAATATTCCTCACTGCTGCCCTCCGTAG-GAGTGTGGACCGGTG--TCAGTTCAGTGT----- 1086

As e bacter chiooi horn
 TCCTG--TCGTGAGATGTT-----GGGTTAAGTCCCGTAACGAGCGCAACCTTGTCTTATGTTACAGCACCT----- 320

Beijerinckia sp. 37L
 TGGCG--TTTTGAAATTT-----CAAACCGGGATCTTT----- 178

Beijerinckia sp. 37L
 TCCTG--TCGTGAGATGTT-----GGGTTAAGTCCCGCAACGAGCGCAACCTTGTCTTATGTTTCAGCATTT----- 1048

Enterobacter doecae OsEp_AN_15A7
 TCCTG--TCGTGAAATGTT-----GGGTTAAGTCCCGCAACGAGCGCAACCTTATCTTTGTTGCACGGGTT----- 1071

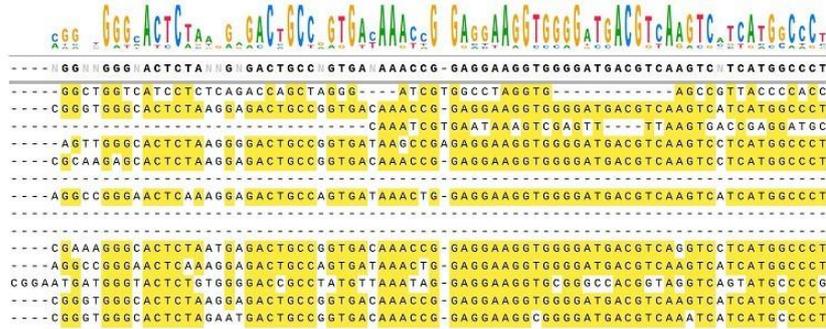
Ideonella sp. URA-8
 TCCTG--TCGTGAGATGTT-----GGGTTAAGTCCCGCAACGAGCGCAACCTTGTCTTATGTTGCTA----- 1104

Klebsiella sp. SXW12
 TCCTG--TCGTGAAATGTT-----GGGTTAAGTCCCGCAACGAGCGCAACCTTATCTTGTGTTGCCAGCGTT----- 1087

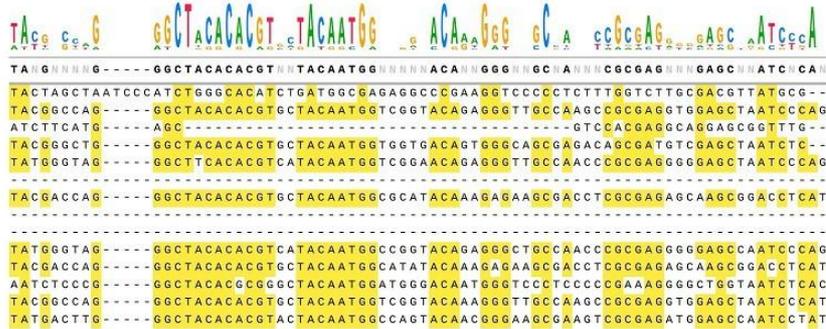
Methanosarcina sp. T4OUC2
 TCCTA--CTGTGAAGCATC-----CTGTTAAGTCCCGCAACGAGCGCAACCTTGTCTTATGTTACCAGCATATTTCT 913

Pseudomonas sp. S23
 TCCTG--TCGTGAGATGTT-----GGGTTAAGTCCCGTAACGAGCGCAACCTTGTCTTATGTTACCAGCACCT----- 1070

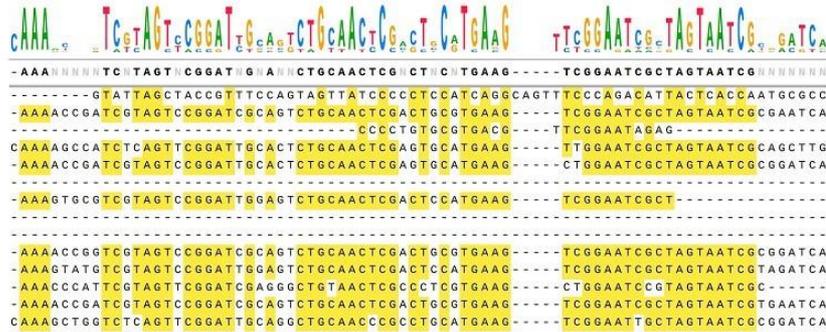
Consensus
Enterobacter sp.GG1
Azo bacter chrooocum Y5G5
Azospirillum lipoferum B7
Beijerinckia sp. 37L
Burkholderia sp.OKR4-1
Clostridium sp.M984-49
Enterobacter doacae OsEp_AN_15A7
Gluconacetobacter sp. 0w26
Herbaspirillum sp.Ah5_2.5
Ideonella sp. URA-8
Klebsiella sp.SXW12
Methanosarcina sp.T40UC2
Pseudomonas sp.S23
Paenibacillus chibensis SK4-3.2



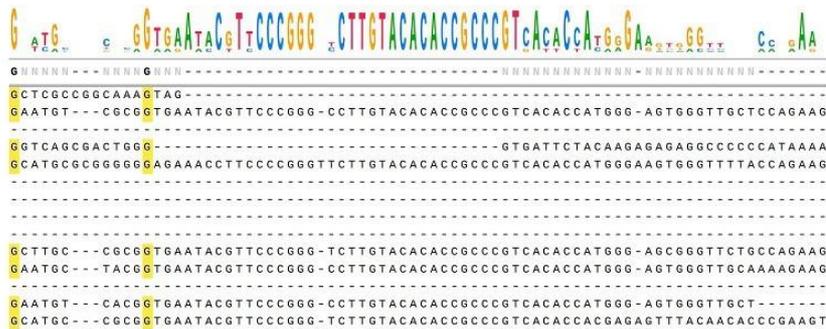
Consensus
Enterobacter sp.GG1
Azo bacter chrooocum Y5G5
Azospirillum lipoferum B7
Beijerinckia sp. 37L
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Clostridium sp.M984-49
Enterobacter doacae OsEp_AN_15A7
Gluconacetobacter sp. 0w26
Herbaspirillum sp.Ah5_2.5
Ideonella sp. URA-8
Klebsiella sp.SXW12
Methanosarcina sp.T40UC2
Pseudomonas sp.S23
Paenibacillus chibensis SK4-3.2

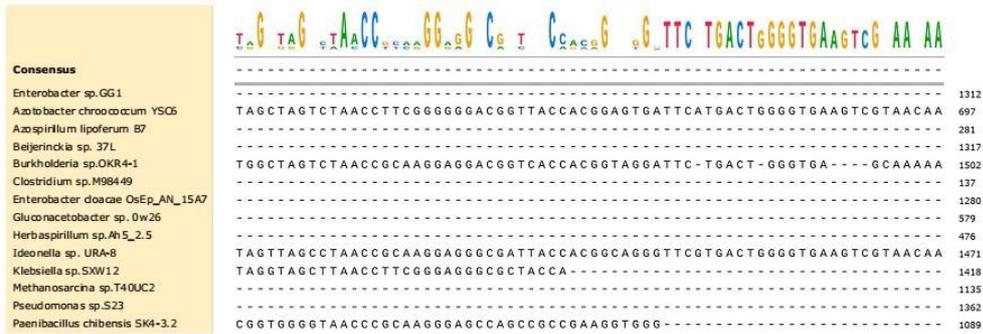


Consensus
Enterobacter sp.GG1
Azo bacter chrooocum Y5G5
Azospirillum lipoferum B7
Beijerinckia sp. 37L
Burkholderia sp.OKR4-1
Clostridium sp.M984-49
Enterobacter doacae OsEp_AN_15A7
Gluconacetobacter sp. 0w26
Herbaspirillum sp.Ah5_2.5
Ideonella sp. URA-8
Klebsiella sp.SXW12
Methanosarcina sp.T40UC2
Pseudomonas sp.S23
Paenibacillus chibensis SK4-3.2



Consensus
Enterobacter sp.GG1
Azo bacter chrooocum Y5G5
Azospirillum lipoferum B7
Beijerinckia sp. 37L
Burkholderia sp.OKR4-1
Clostridium sp.M984-49
Enterobacter doacae OsEp_AN_15A7
Gluconacetobacter sp. 0w26
Herbaspirillum sp.Ah5_2.5
Ideonella sp. URA-8
Klebsiella sp.SXW12
Methanosarcina sp.T40UC2
Pseudomonas sp.S23
Paenibacillus chibensis SK4-3.2





Supplementary Table 1: Comparative data analysis of nitrogen fixing microbes with newly isolates *Enterobacter* sp. GG1

Name of organism	Comparative features	Reference
<i>Enterobacter</i> sp. GG1	Effective nitrogen fixation, growth rate, variability very high, non-pathogenic, effective efficacy result show in saline soil	Current study
<i>Pseudomonas aeruginosa</i>	Nitrogen fixation rate high but pathogenicity very high	[28]
<i>Frankia</i> sp.	Nitrogen fixation rate high but that become infected the actinorhizal plants	[52]
<i>Corynebacterium</i> sp.	Nitrogenase activity and viability little bet low	[53]
<i>Clostridium botulinum</i>	Nitrogen fixation rate high but pathogenicity very high, growth rates declined with and increasing saline stress condition	[29]
<i>Azotobacter</i> sp.	Nitrogen fixation rate very high but high salt concentration and temperature	[54]
<i>Anabaena</i> sp.	Nitrogen fixation rate very high but growth rate is very low compare to bacterial growth	[55]