

Genes on +/- strand:

CDS:

RNA features:

Misc features:

Core Genome:

*Lysinibacillus*_sp_CD3_6_1_contig_1

*Lysinibacillus*_sp_JNUCC_52_CP065546

GC-content (1.000 bp window):

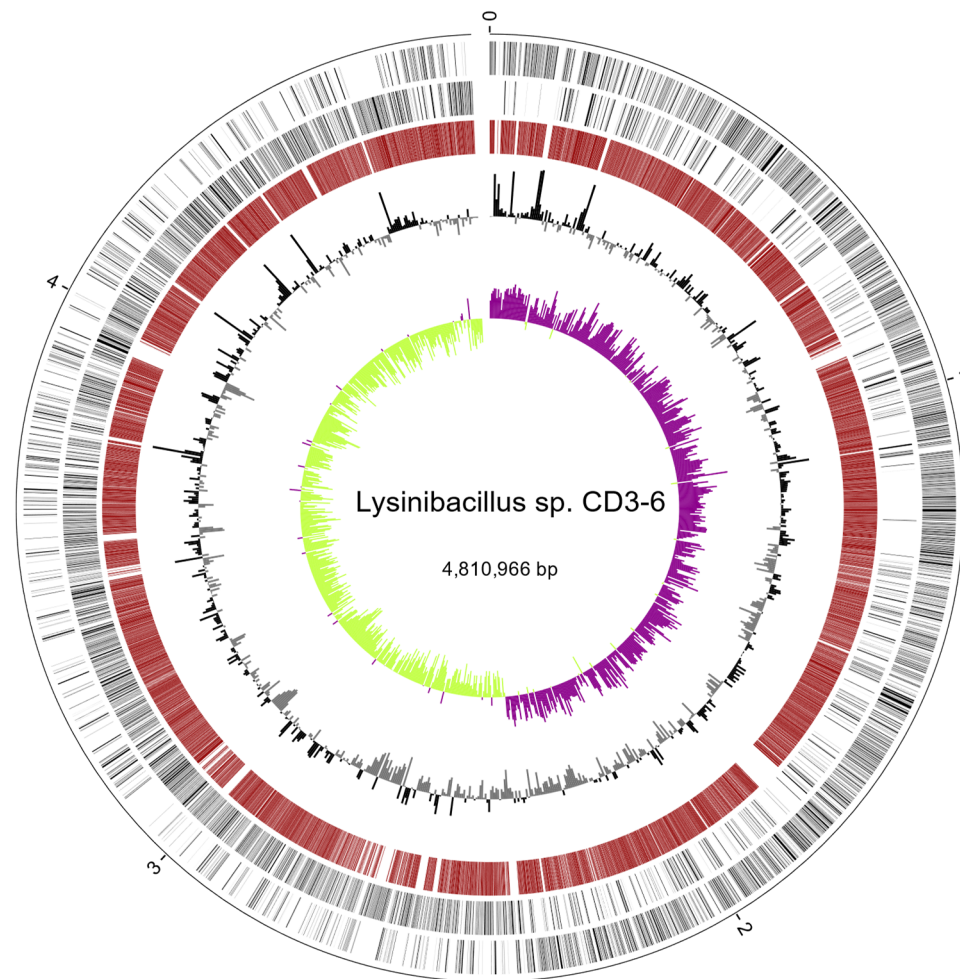
above mean:

below mean:

GC Skew $[(G - C)/(G + C)]$ (1.000 bp window):

above mean:

below mean:



Supplementary Figure S1: Circular plot of the genome of *Lysinibacillus* sp. CD3-6 generated with BioCircos (Cui et al. 2016). The genome of CD3-6 was computed against *Lysinibacillus* sp. JNUCC-52. Core genome genes shared by *Lysinibacillus* sp. CD3-6 and *Lysinibacillus* sp. JNUCC-52 are indicated in brown color in the third circle.

Genes on +/- strand:

CDS:

RNA features:

Misc features:

Core Genome:

Brevibacillus_sp_HD3_3_A_BHD33A_1
 Brevibacillus_parabrevis_strain_179_C7_2_HS_NZ_JAHYZ01000009
 Brevibacillus_parabrevis_strain_CN1_NZ_LQQV01000136
 Brevibacillus_parabrevis_strain_NBRC_12334_NZ_BJMH01000119
 Brevibacillus_parabrevis_strain_NRR_L_NRS_605_NZ_RHHV01000009
 Brevibacillus_sp_HD1_4A_NZ_JABSUW010000099

Pairwise alignment Brevibacillus_sp_HD3_3_A_BHD33A_1 with:

Brevibacillus_parabrevis_strain_179_C7_2_HS_NZ_JAHYZ01000009
 Brevibacillus_parabrevis_strain_CN1_NZ_LQQV01000136
 Brevibacillus_parabrevis_strain_NBRC_12334_NZ_BJMH01000119
 Brevibacillus_parabrevis_strain_NRR_L_NRS_605_NZ_RHHV01000009
 Brevibacillus_sp_HD1_4A_NZ_JABSUW010000099

GC-content (1,000 bp window):

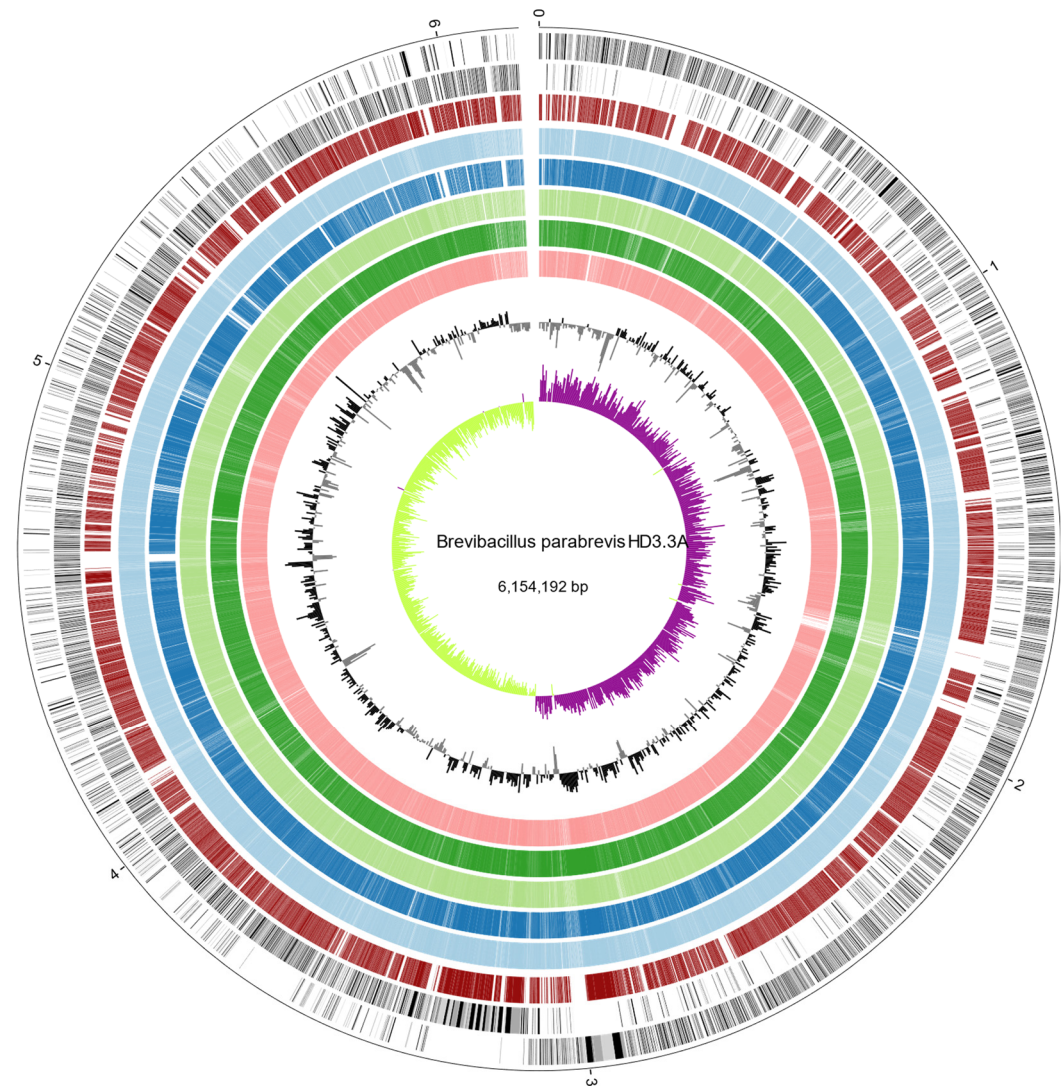
above mean:

below mean:

GC Skew $[(G - C)/(G + C)]$ (1,000 bp window):

above mean:

below mean:



Supplementary Figure S2: Circular plot of the genome of *Brevibacillus parabrevis* HD3.3A generated with BioCircos (Cui et al. 2016). HD3.3A was used as reference for computing the core genome against representatives of the *Brevibacillus*-A5 branch (clusters 25-27, see section 3.2). Genes of the core genome are presented in the third circle.

Genes on +/- strand:

CDS:

RNA features:

Misc features:

Core Genome:

Brevibacillus_sp_DP1_3_A_BDP13A_1
 Brevibacillus_sp_BC25_NZ_AKIX01000140
 Brevibacillus_sp_MS2_2_NZ_JABSUZ010000009
 Brevibacillus_sp_Leaf182_NZ_LMPN02000032
 Brevibacillus_brevis_strain_DZQ7_NZ_CP030117

Pairwise alignment Brevibacillus_sp_DP1_3_A_BDP13A_1 with:

Brevibacillus_sp_BC25_NZ_AKIX01000140
 Brevibacillus_sp_MS2_2_NZ_JABSUZ010000009
 Brevibacillus_sp_Leaf182_NZ_LMPN02000032
 Brevibacillus_brevis_strain_DZQ7_NZ_CP030117

GC-content (1.000 bp window):

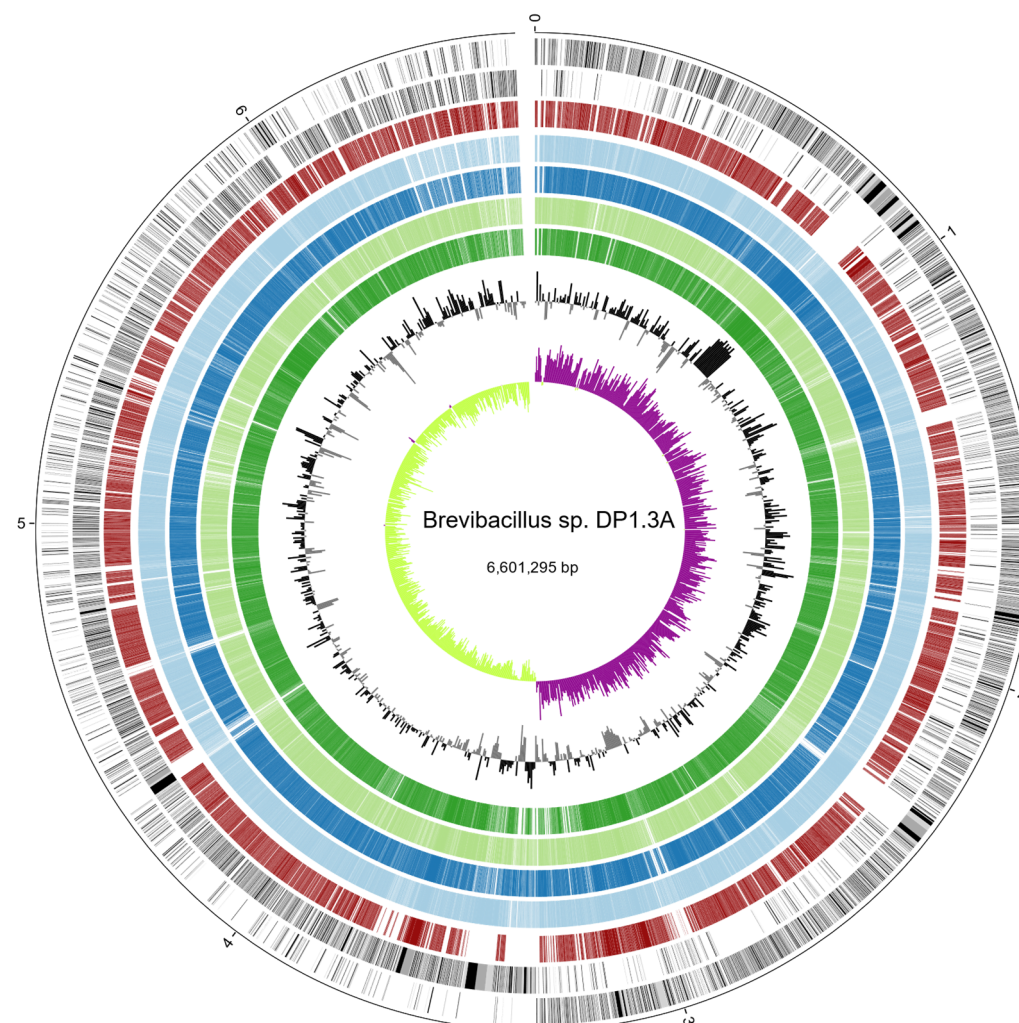
above mean:

below mean:

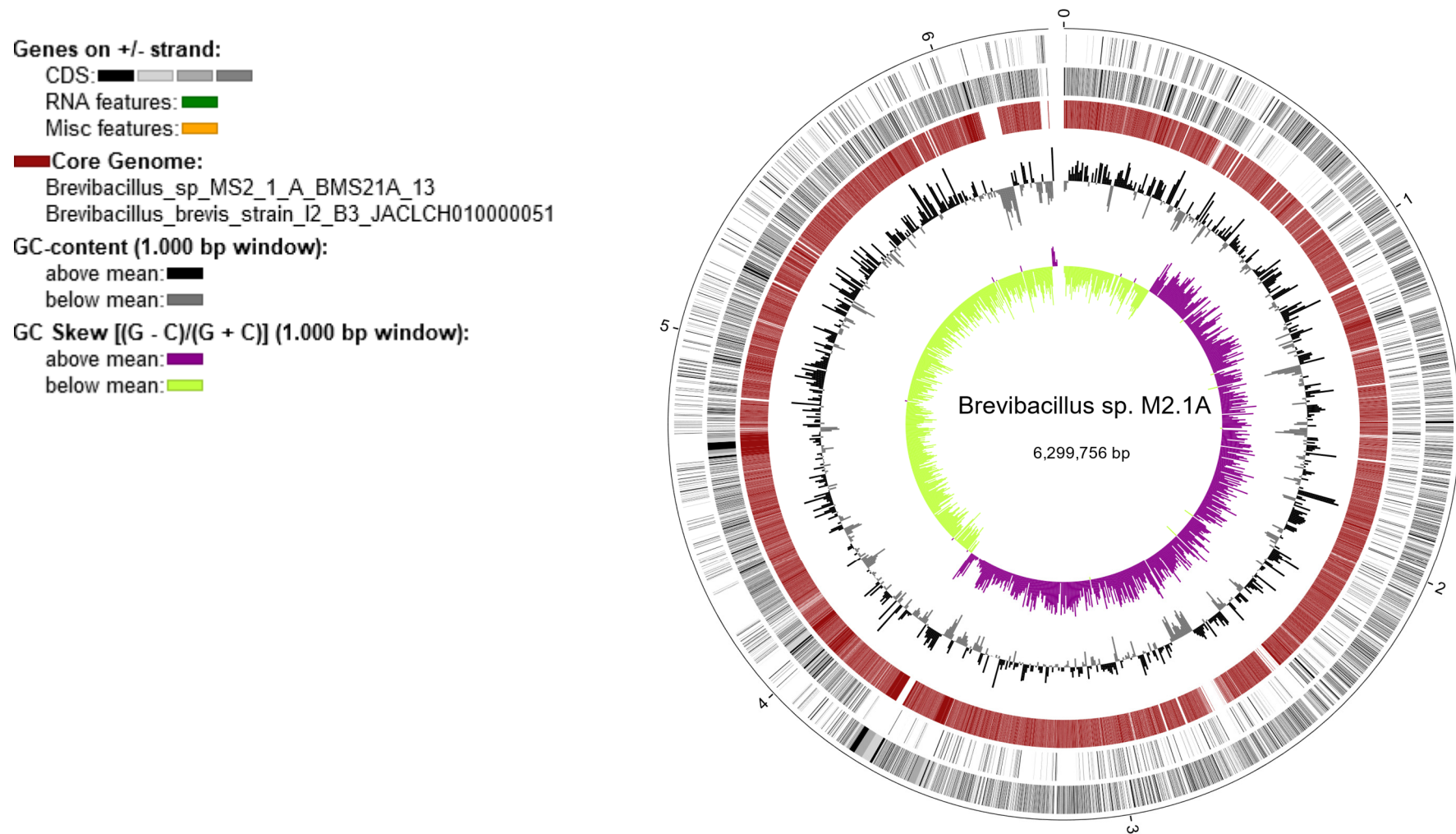
GC Skew $[(G - C)/(G + C)]$ (1.000 bp window):

above mean:

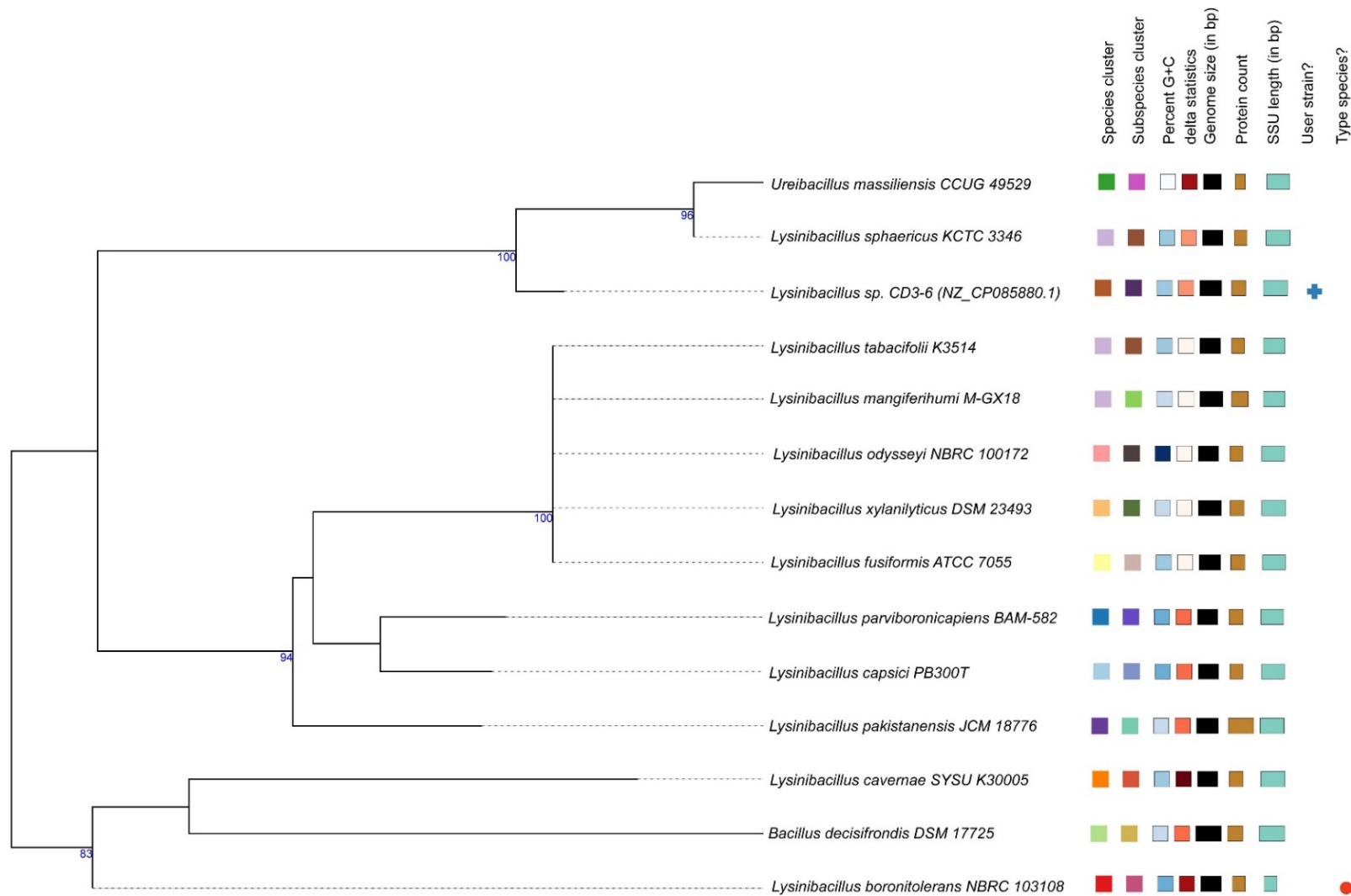
below mean:



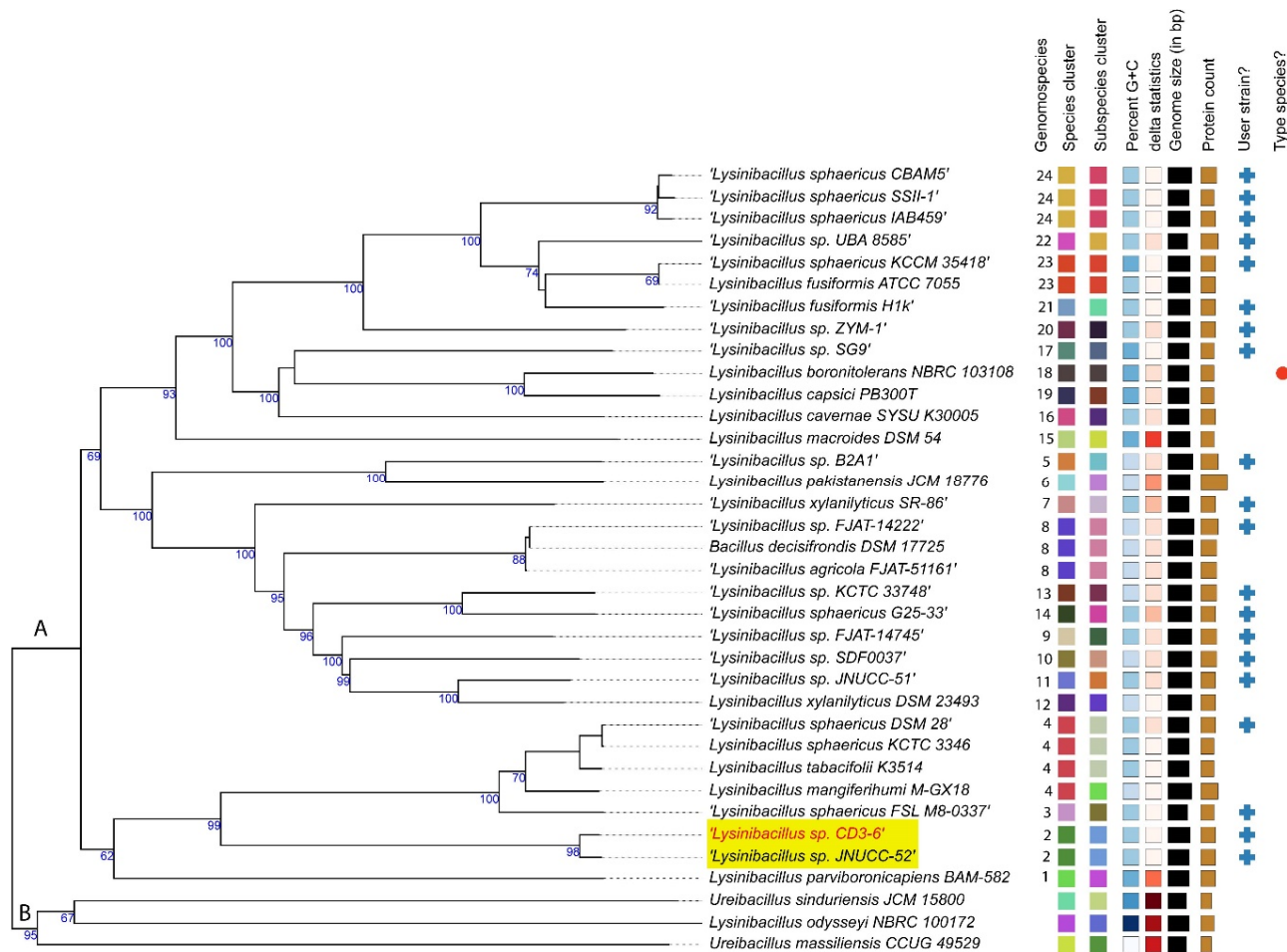
Supplementary Figure S3: Circular plot of the genome of *Brevibacillus* sp. DP1.3A generated with BioCircos (Cui et al. 2016). The DP1.3A genome was computed against selected members of the *Brevibacillus*-A6 branch (clusters 28-35). Homologous regions representing the core genome are visualized in the third circle.



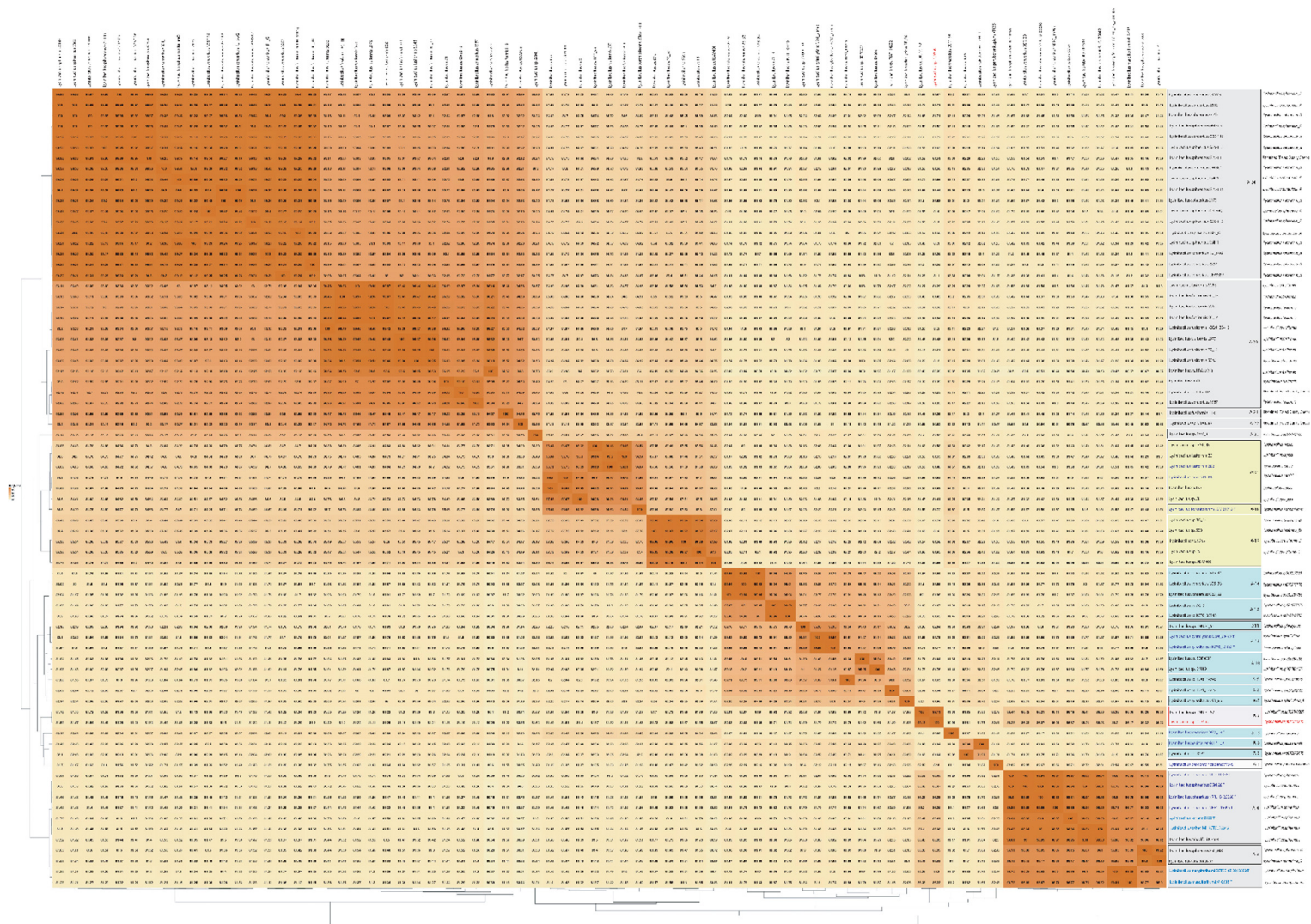
Supplementary Figure S4: Circular plot of the genome of *Brevibacillus* sp. M2.1A generated with BioCircos. The genome of M2.1A was computed against *B. brevis* 12B3. Core genome genes are indicated in brown color in the third circle



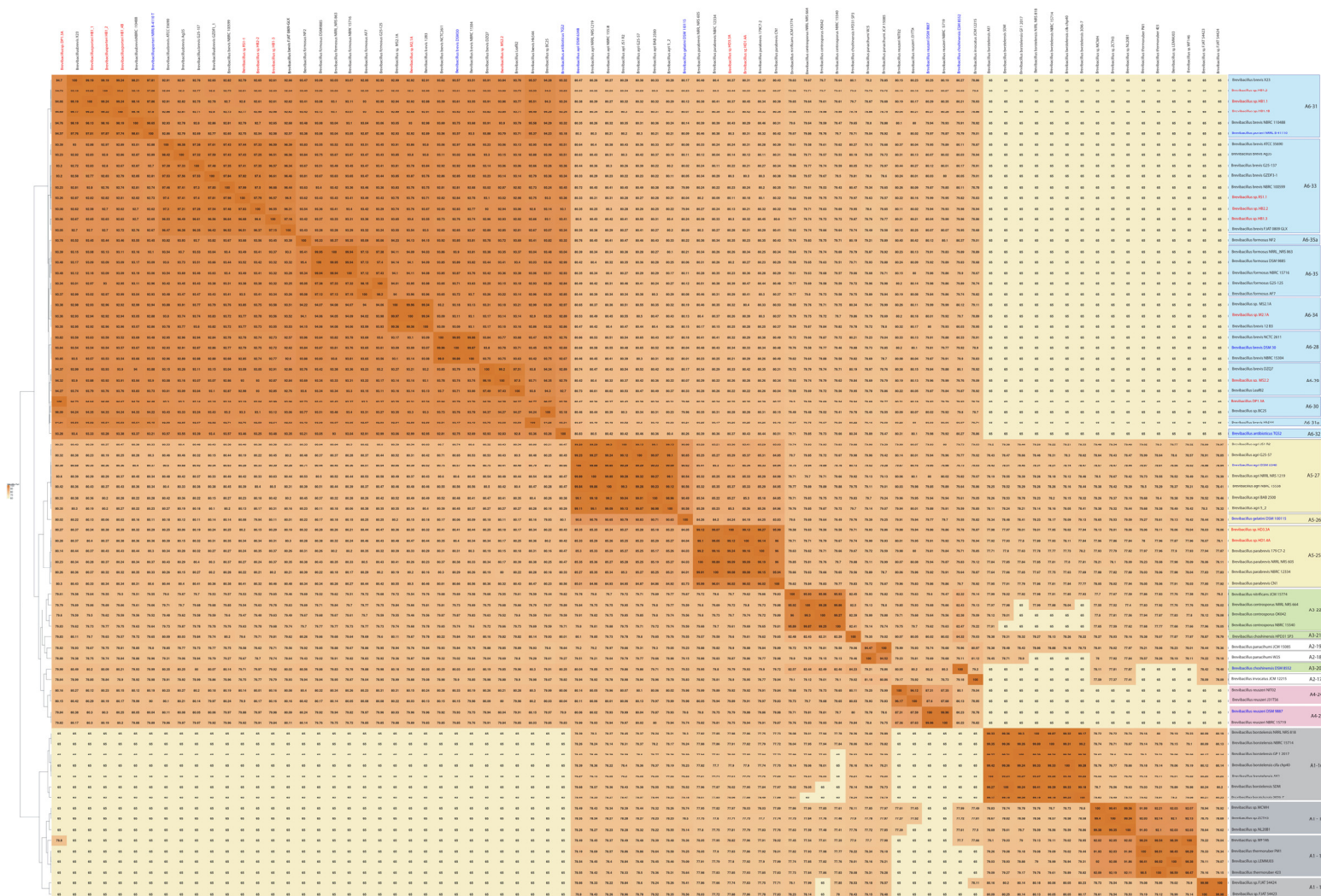
Supplementary Figure S5: GBDP tree (16S rDNA gene sequence based) with *Lysinibacillus* sp. CD3-6. Tree inferred with FastME 2.1.6.1 from GBDP distances calculated from 16S rDNA gene sequences. The branch lengths are scaled in terms of GBDP distance formula d_5 . The numbers above branches are GBDP pseudo-bootstrap support values > 60 % from 100 replications, with an average branch support of 54.8 %. The tree was rooted at the midpoint.



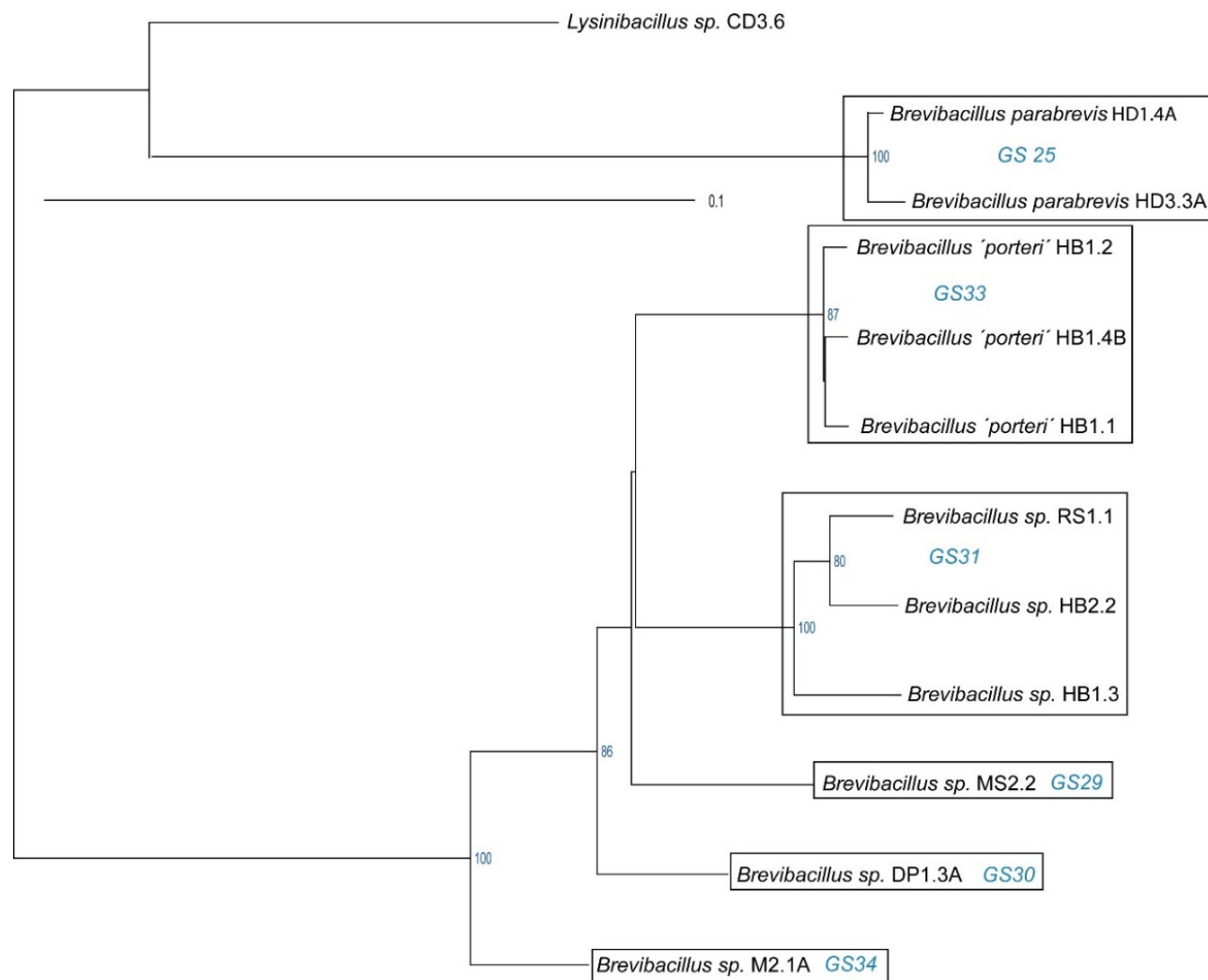
Supplementary Figure S6: *Lysinibacillus* tree inferred with FastME 2.1.6.1 from GBDP distances calculated from 37 entries. Clustering of the *Lysinibacillus* strains revealed 28 species and 29 subspecies clusters. Two of them could not be assigned to known species. *Lysinibacillus* CD3-6 was assigned to form together with *Lysinibacillus* sp. JNUCC-52 a novel species cluster. The branch lengths are scaled in terms of GBDP distance formula d_5 . The numbers above branches are GBDP pseudo-bootstrap support values > 60 % from 100 replications, with an average branch support of 93.5 %. The tree was rooted at the midpoint.



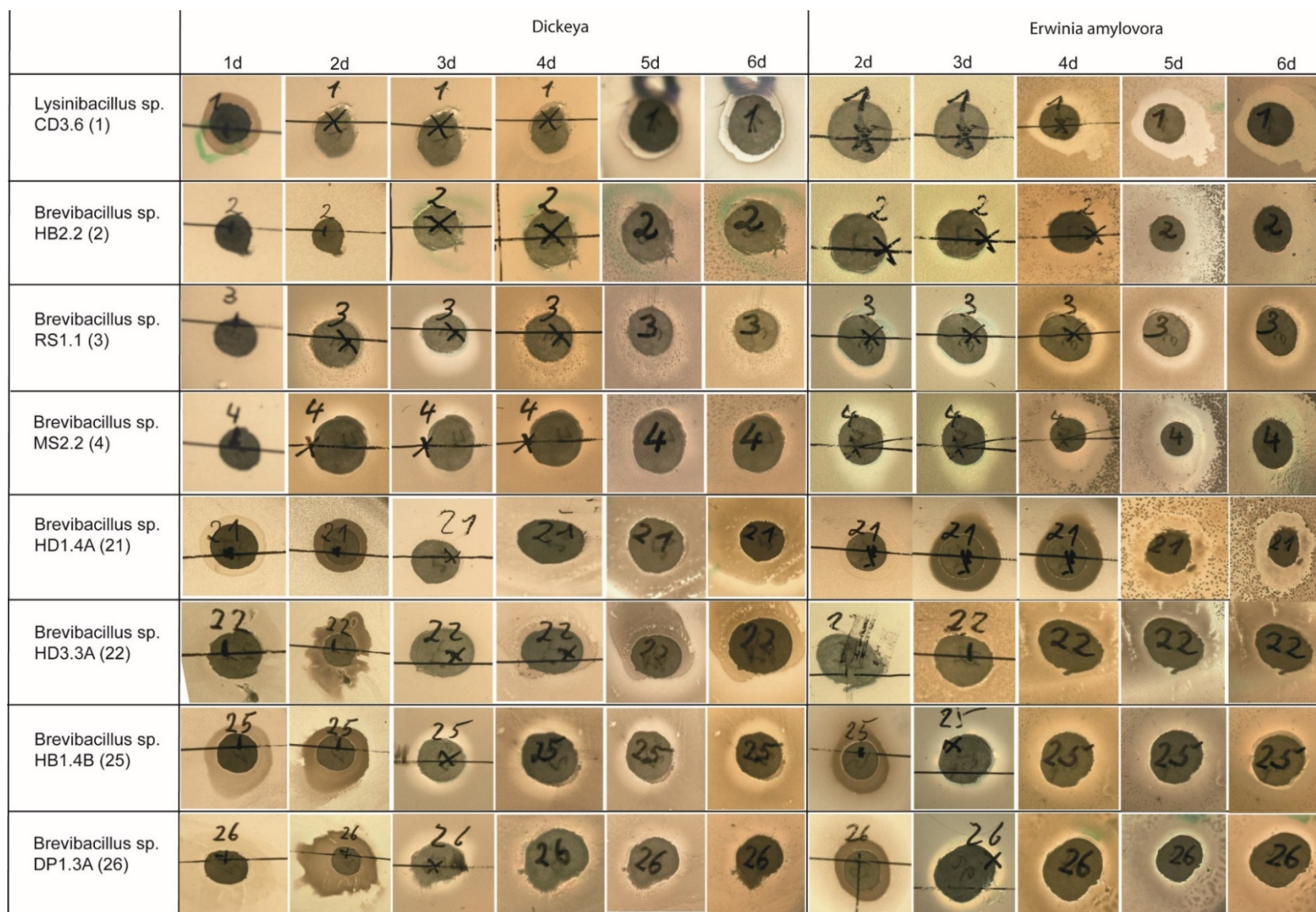
Supplementary Figure S7. Heat Map of the FastANI-Matrix [23] of group A *Lysinibacillus* genomes. *Lysinibacillus* sp. CD3-6 (labelled in red letters) isolated from Vietnamese crop plants formed together with JNUCC-52 a separate genomospecies. Average nucleotide identity and aligned nucleotides [%] were determined using JSpecies <https://jspecies.ribohost.com/> [42]. For better resolution use the separate png file of Suppl. Fig. S7.



Supplementary Figure S8. Heat Map of the FastANI-Matrix [23] of *Brevibacillus* strains isolated from Vietnamese crop plants. Average nucleotide identity and aligned nucleotides [%] were determined using JSpecies <https://jspecies.ribohost.com/> [42]. For better resolution use the separate png file of Suppl. Fig. S8.

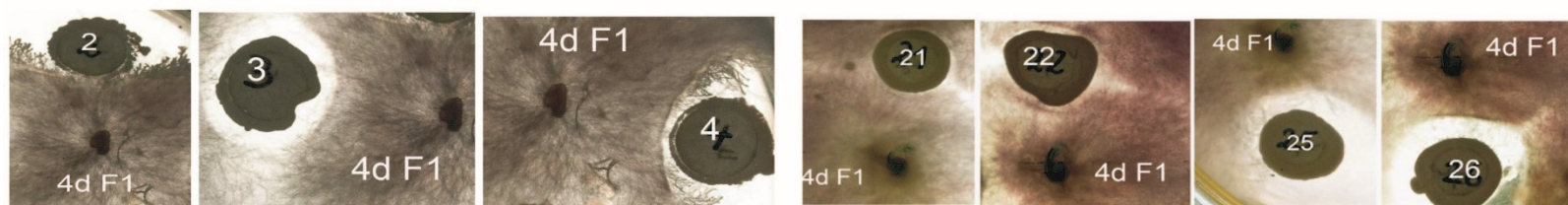


Supplementary Figure S9: Phylogenetic tree constructed with the Vietnamese *Brevibacillus* isolates. The corresponding genospecies clusters are indicated. *Lysinibacillus* sp. CD3-6 was used as outgroup.



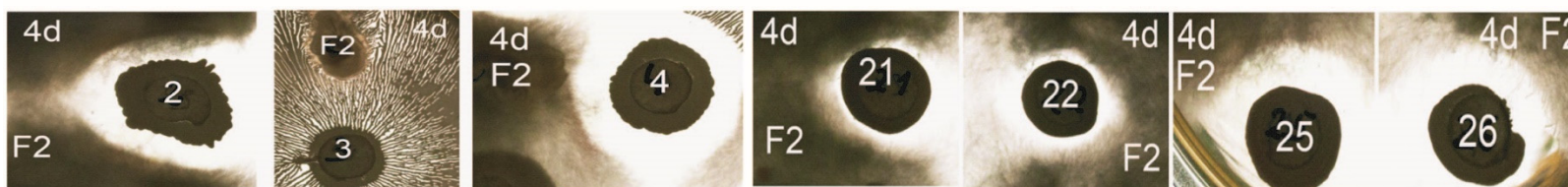
Supplementary Figure S10: In vitro assay of biocontrol action of *Lysinibacillus* and *Brevibacillus* strains against *Dickeya solani* and *Erwinia amylovora*.

A



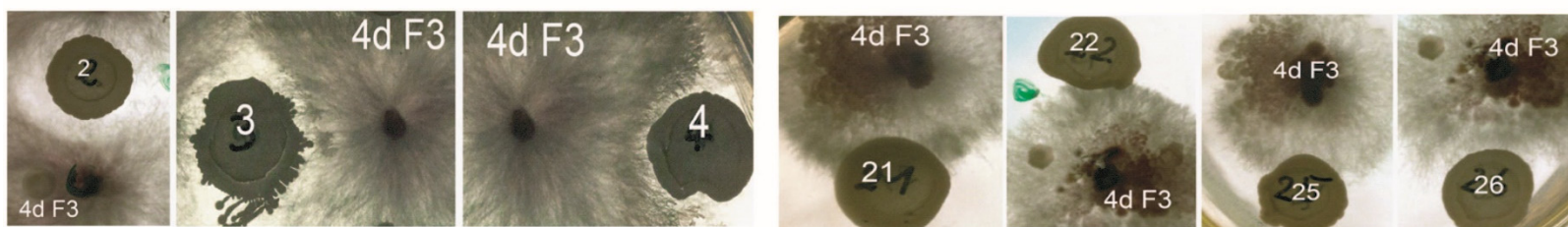
Fusarium culmorum

B



Fusarium poae

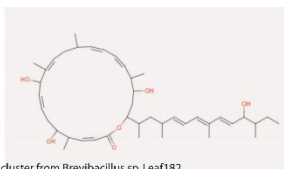
C



Fusarium graminearum

Supplementary Figure S11: Antagonistic action of *Brevibacillus* sp. HB2.2 (2), *Brevibacillus* sp. RS1.1 (3), *Brevibacillus* sp. MS2.2 (4), *B. parabrevis* HD1.4A (21), *Brevibacillus parabrevis* (22), *Brevibacillus porteri* HB1.4B (25), and *Brevibacillus* DP1.3A (26) against *Fusarium culmorum* (A), *Fusarium poae* (B), and *Fusarium graminearum* (C) after 4 days incubation at 27°C.

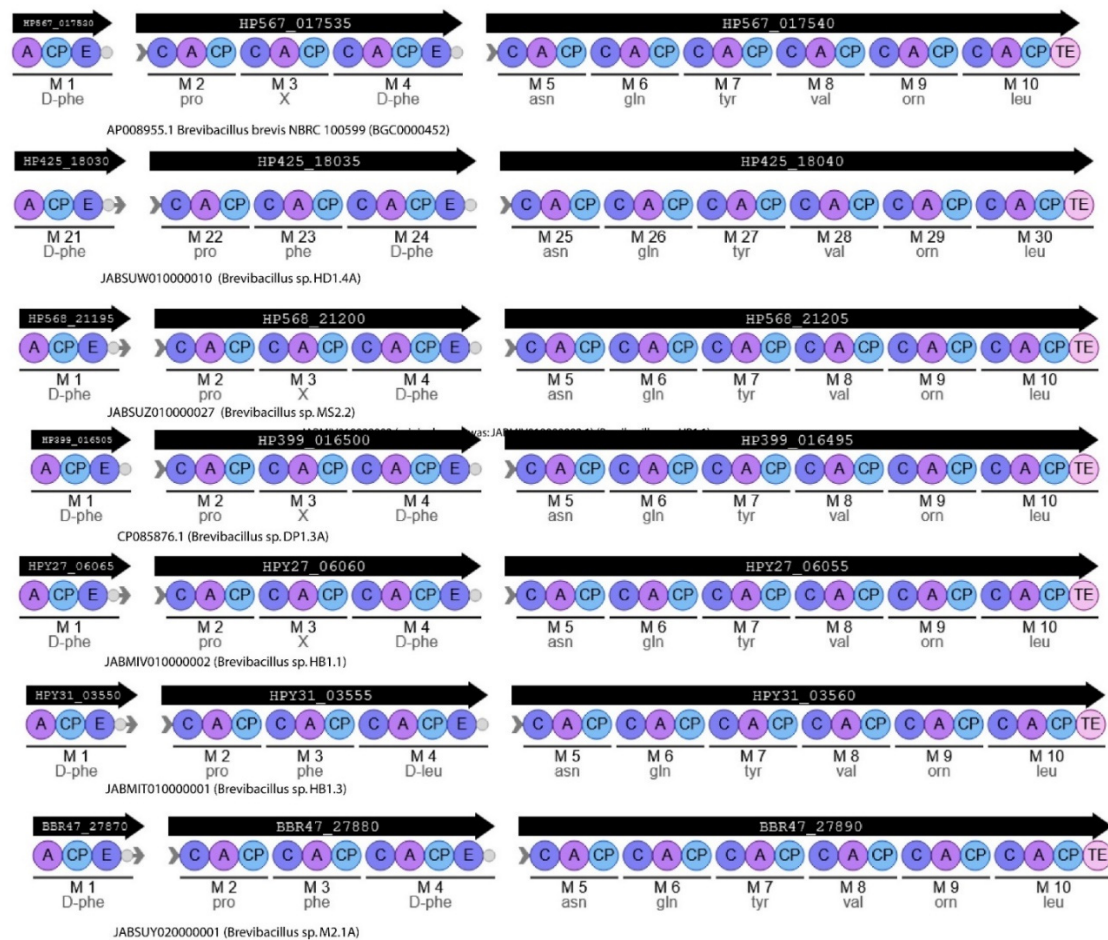
Macrobrevin



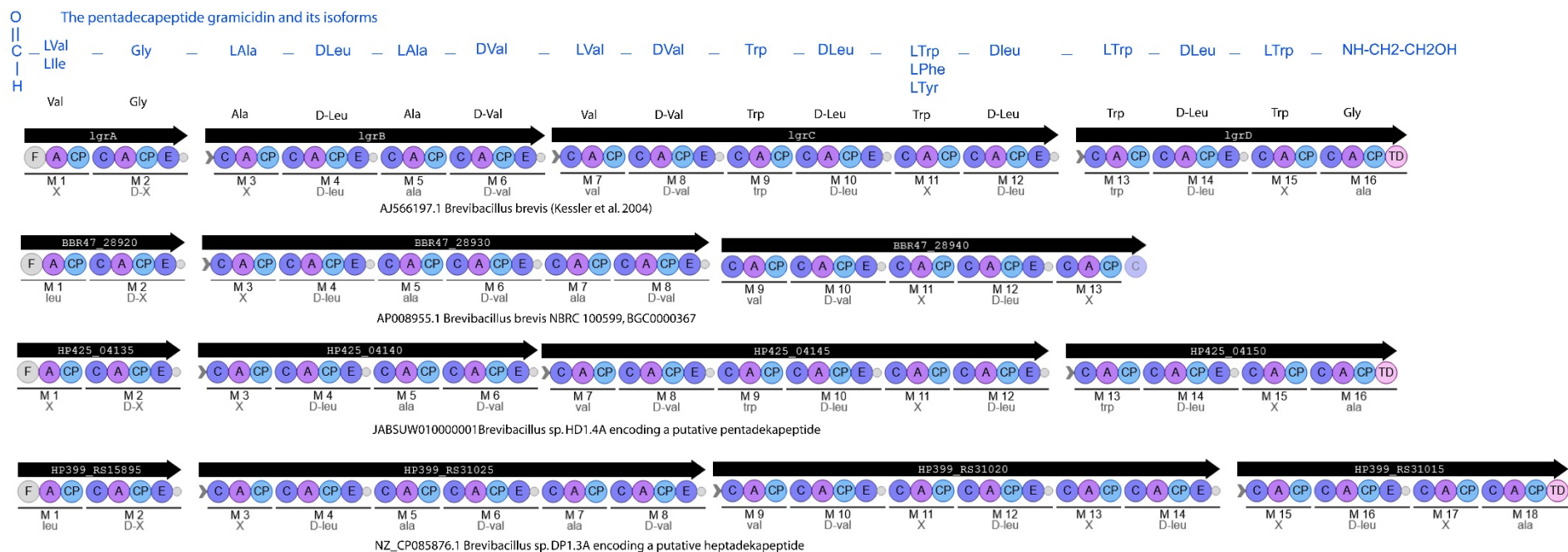
BGC0001470:macrobrevin biosynthetic gene cluster from *Brevibacillus* sp. Leaf182



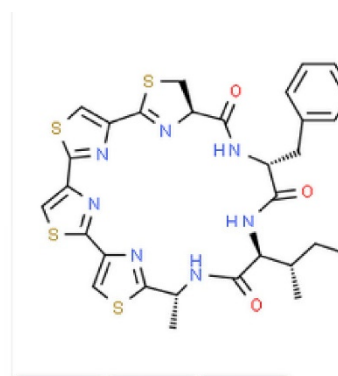
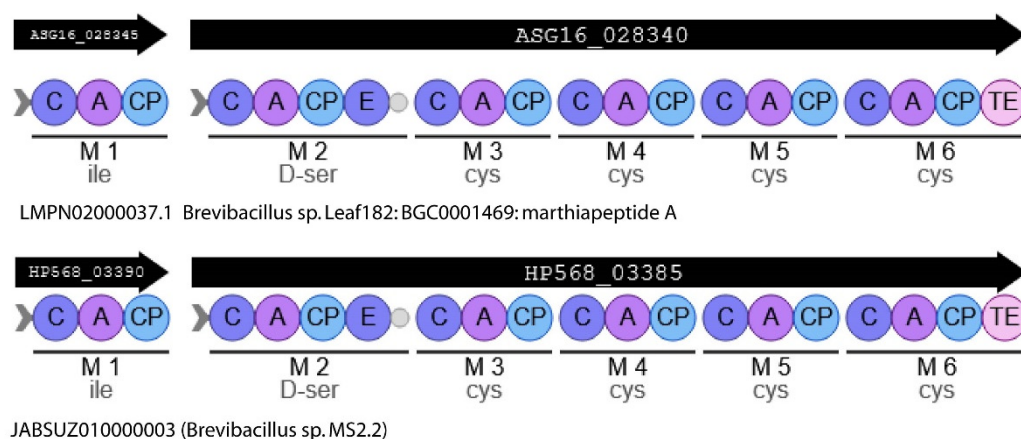
Supplementary Figure S12: Modular PKS in *Brevibacilli*: The Macrobrevin variants harbored slight differences compared to *Brevibacillus* sp. leaf 182 (BGC0001470). NRPS/PKS domains are indicated by filled circles. Fade circles indicate inactive domains. Crosses mean that the corresponding domain is not complete. CP: condensation domain, DH: dehydratase domain, KS: ketosynthase domain, KR: ketoreductase, TE: thioesterase.



Supplemental Figure S13: NRP: Gene clusters similar to Tyrocidine (BGC0000452) present in *Brevibacillus parabrevis* HD1.4A (GS 25), *Brevibacillus* sp. MS2.2, DP1.3A, HB1.1, and M2.1A.



Supplemental Figure S14: NRP: Gene clusters similar to gramicidin (BGC0000367) and the gramicidin gene cluster in *Brevibacillus brevis* (AJ566197) were detected in *Brevibacillus parabrevis* HD1.4A (GS 25), and *Brevibacillus* sp. DP1.3A. The pentadecapeptides gramicidin A, B, C are shown above in blue letters. The complete gene clusters detected in *Brevibacillus brevis* (AJ566197), *Brevibacillus parabrevis* HD1.4A (JABSUW010000001), and *Brevibacillus* sp. DP1.3A (CP085876) were characterized by a formylation (F) domain in the first module (*lgrA*) and a final reductase (TD) domain in the last module (*lgrD*).

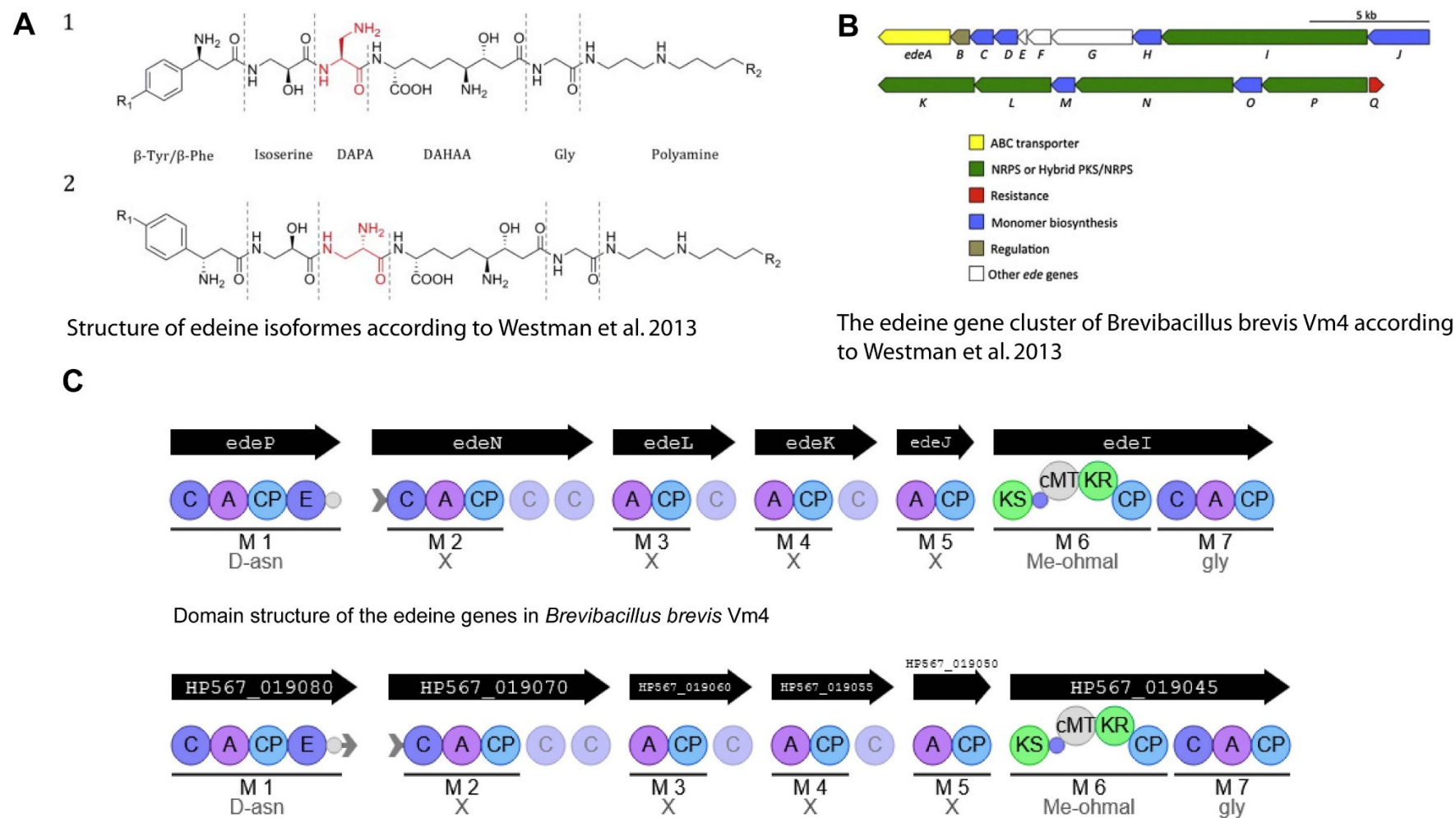


marthiapeptide A

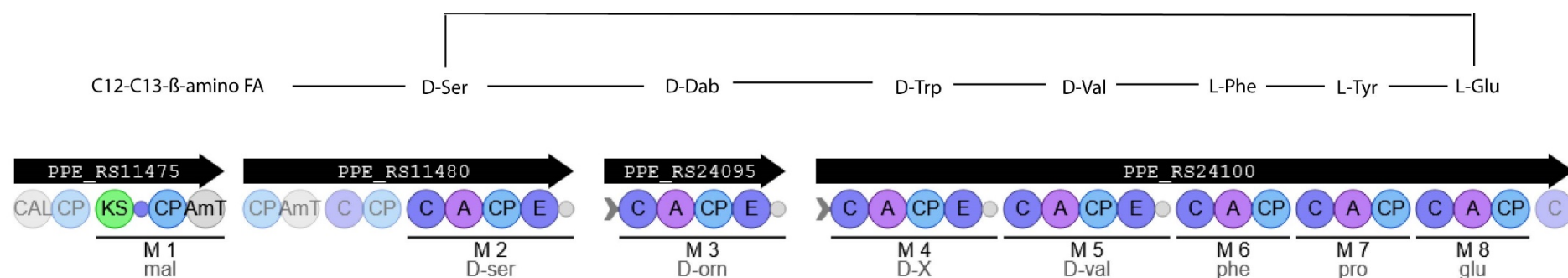
Molecular Formula	C ₃₀ H ₃₁ N ₇ O ₃ S ₄
Average mass	665.872 Da
Monoisotopic mass	665.137146 Da
ChemSpider ID	28536074

- 5 of 5 defined stereocentres

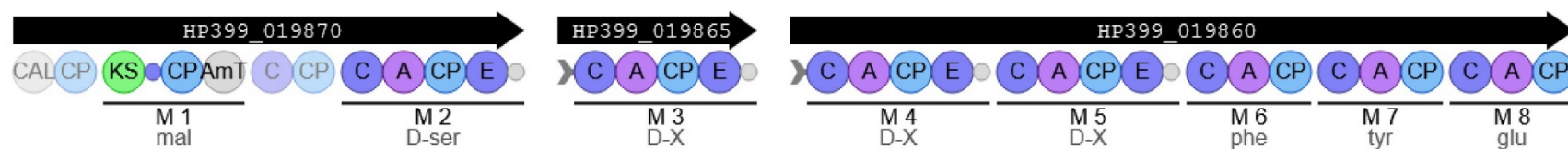
Supplemental Figure S15: NRP: Gene cluster similar to marthiapeptide A (BGC0001469) was detected in *Brevibacillus* sp. MS2.2. The structure of marthiapeptide A was taken from ChemSpider (ID 28536074)



Supplementary Figure S16: Modular PKS-NRP hybrids. A: Structure of edeine isoforms according to Westman et al. 2013. B: The edeine gene cluster of *Brevibacillus brevis* Vm4. C: Prediction of the domain structure in the *Brevibacillus* M2.1A gene cluster similar to the edeine gene cluster of *Brevibacillus brevis* Vm4 in Vietnamese *Brevibacillus* genomes by **antiSMASH 6.0**.

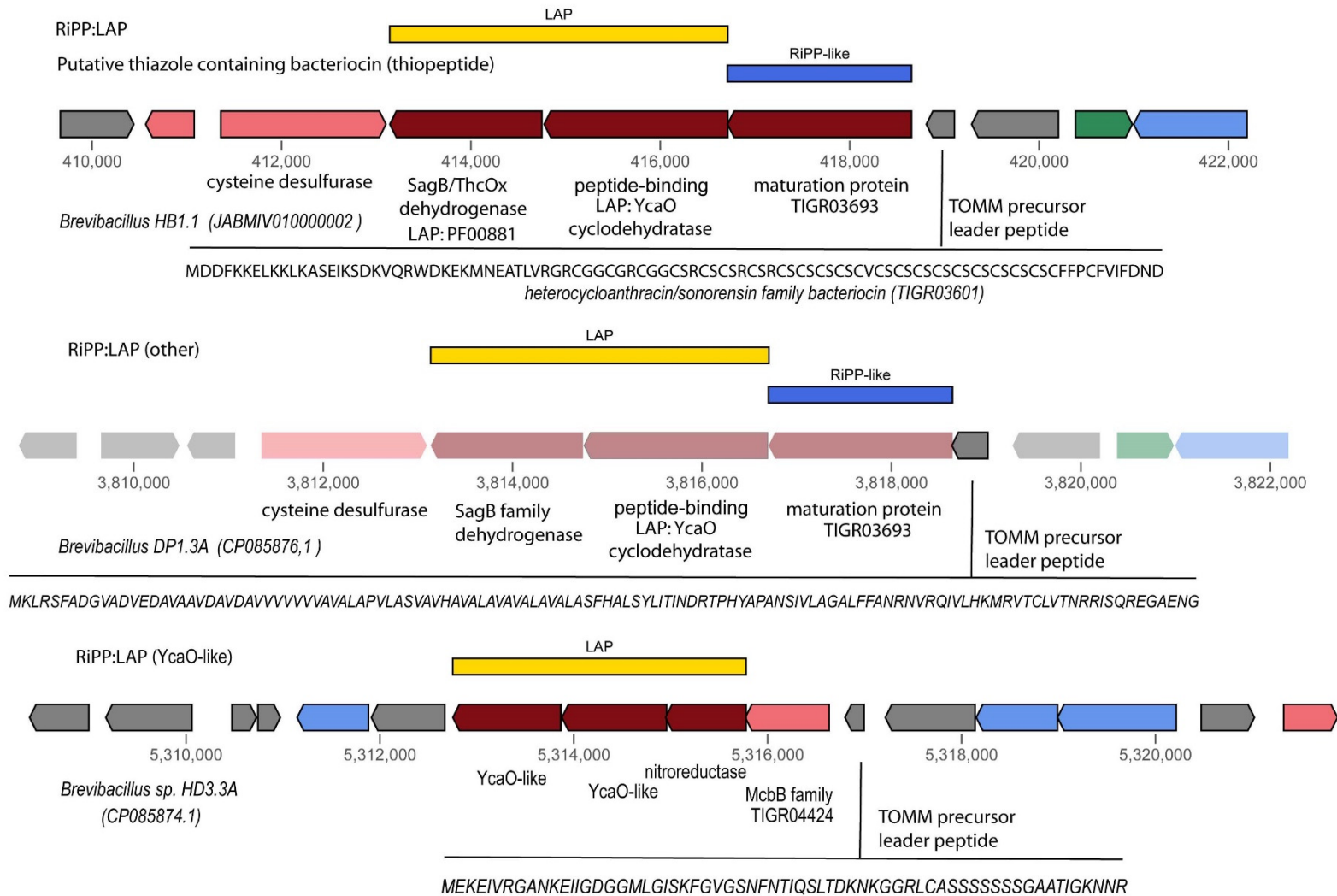


Domain structure of the Paenilipoheptin gene cluster of *Paenibacillus polymyxa* E681 (BGC0001728) revealed by antiSMASH 6.0 analysis

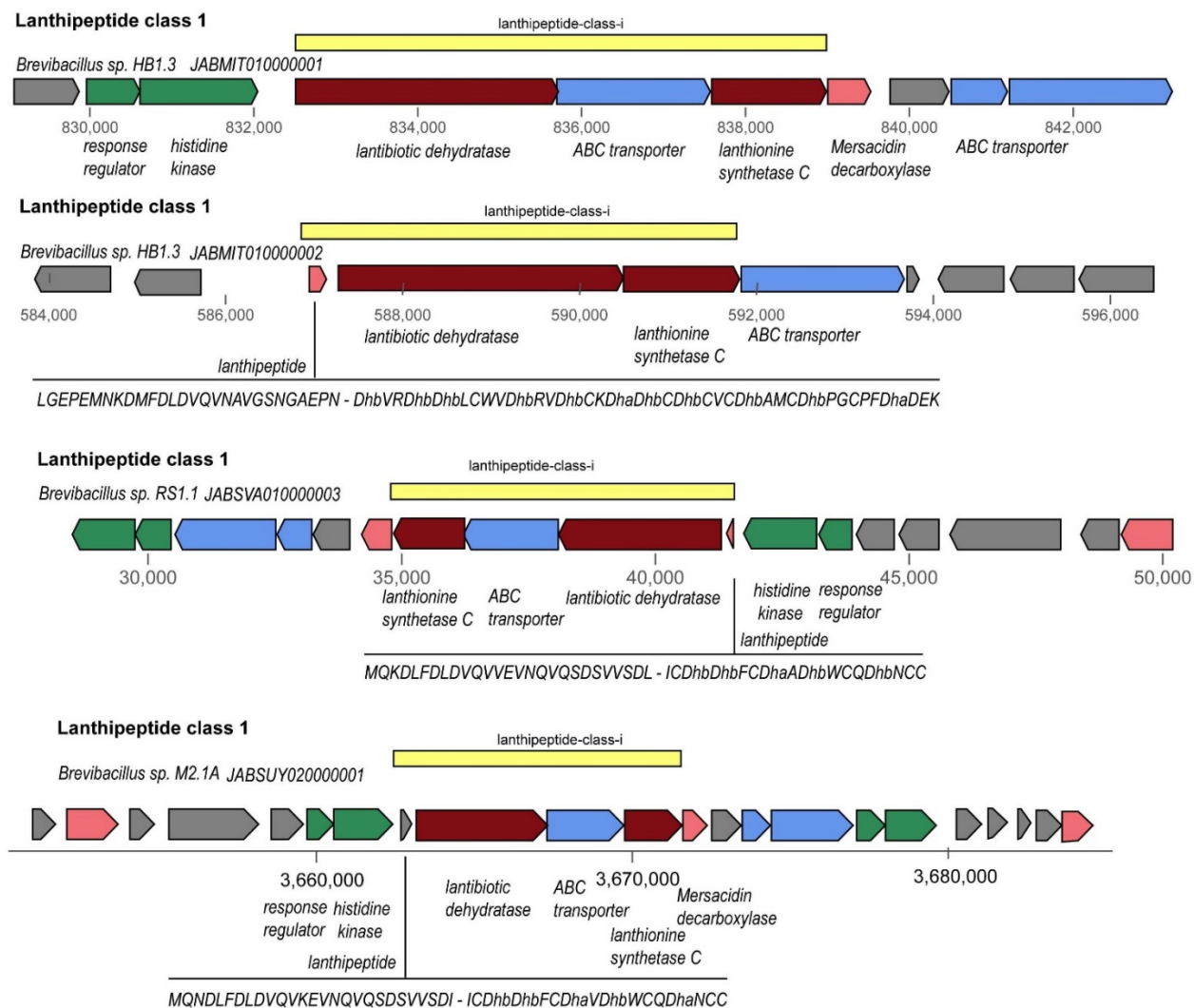


Domain structure of a gene cluster in *Brevibacillus* sp. DP1.3A similar to the Paenilipoheptin gene cluster of *Paenibacillus polymyxa* E681 (BGC0001728)

Supplementary Figure S17: Modular PKS-NRP hybrids. Gene cluster similar to the gene cluster encoding the synthesis of the cyclic lipopeptide paenilipoheptin from *Paenibacillus polymyxa* E681 (BGC0001728) was detected in *Brevibacillus* sp. DP1.3A by antiSMASH 6.0 analysis. The sequence of paenilipoheptin is shown on the upper part (Vater et al. 2018).



Supplemental Figure S18: RiPP-LAP in *Brevibacillus* sp. HB1.1, DP1.3A, and HD3.3A

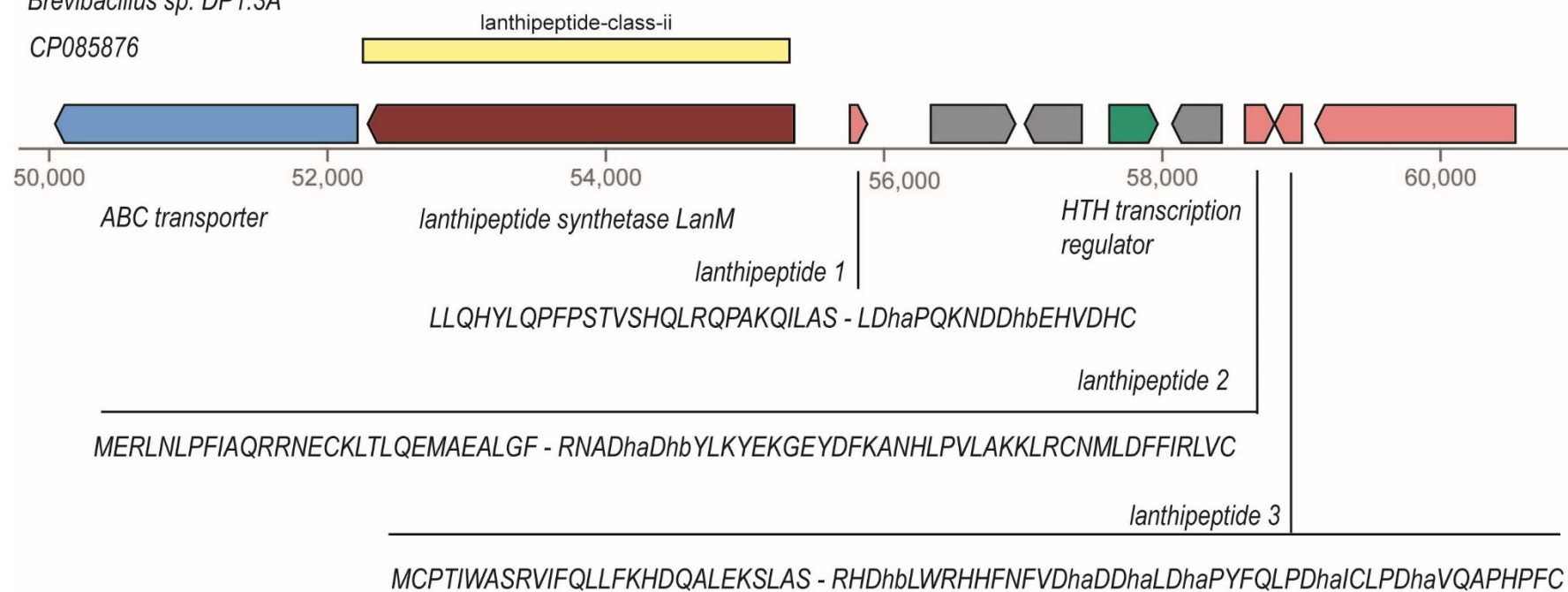


Supplementary Figure S19: Class 1 lanthipeptides detected in *Brevibacillus* sp. HB1.3, *Brevibacillus* sp. RS1.1, and *Brevibacillus* sp. M2.1A

RiPP_Lanthipeptide class ii

Brevibacillus sp. DP1.3A

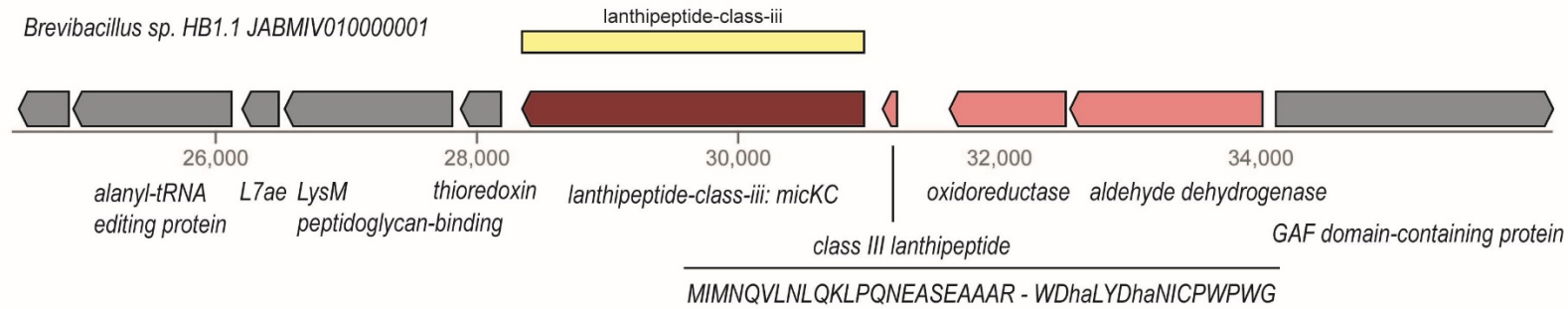
CP085876



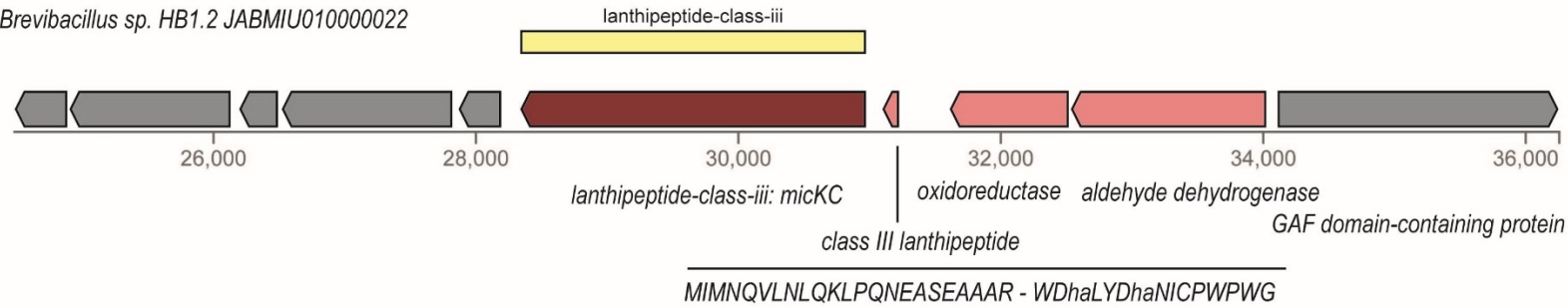
Supplemental Figure S20: RiPP-Lanthipeptides class 2 in *Brevibacillus* sp. DP1.3A

RiPP Lanthipeptide class iii

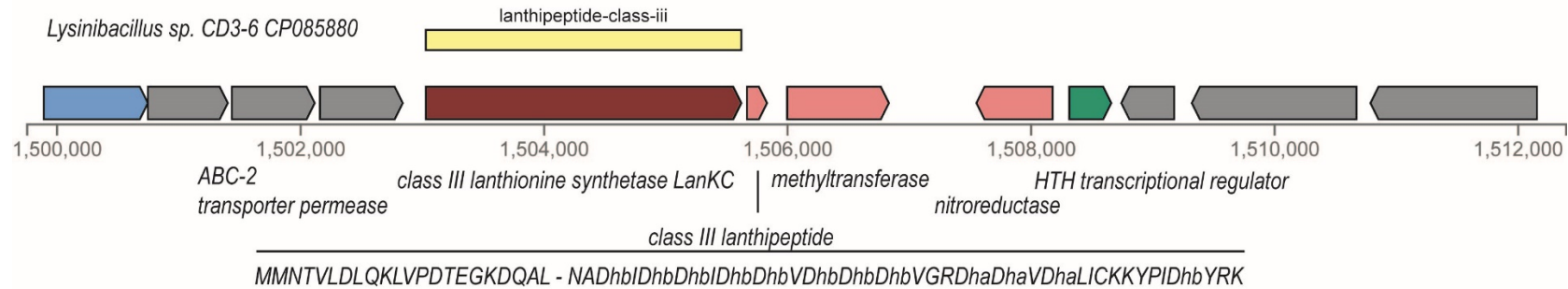
Brevibacillus sp. HB1.1 JABMIV010000001



Brevibacillus sp. HB1.2 JABMIU010000022



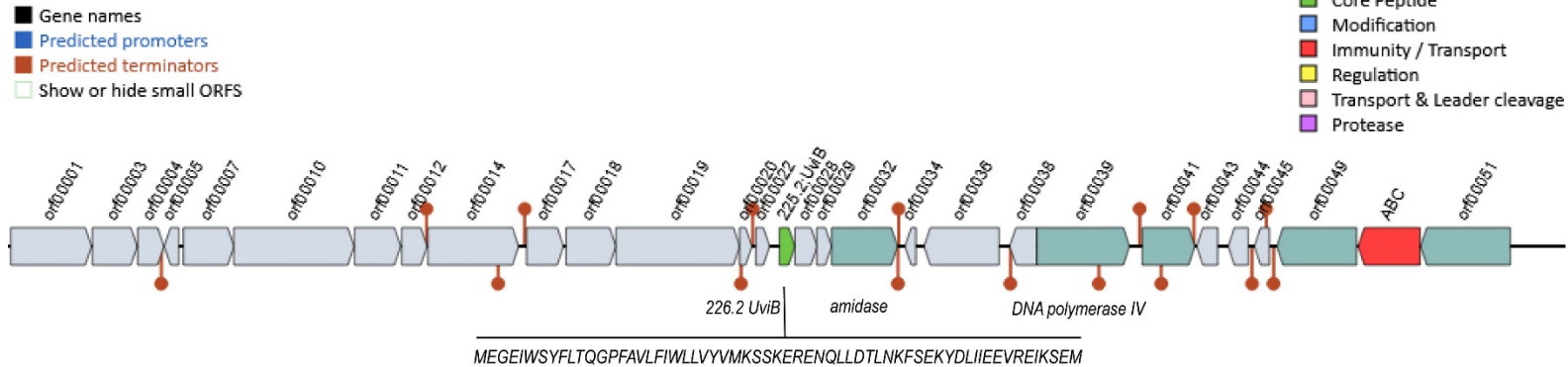
Lysinibacillus sp. CD3-6 CP085880



Supplemental Figure S21: RiPP lanthipeptides class 3 in *Brevibacillus porteri* HB1.1, HB1.2, and *Lysinibacillus* CD3-6

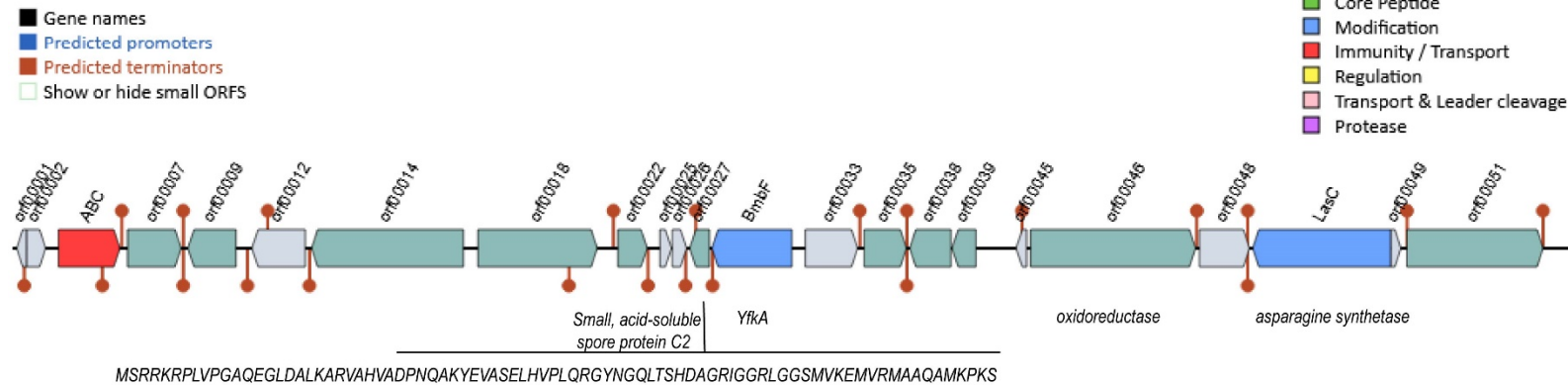
UViB

JABSUW010000001.1Brevibacilluspp.HD1.4A1.fasta AOI_01

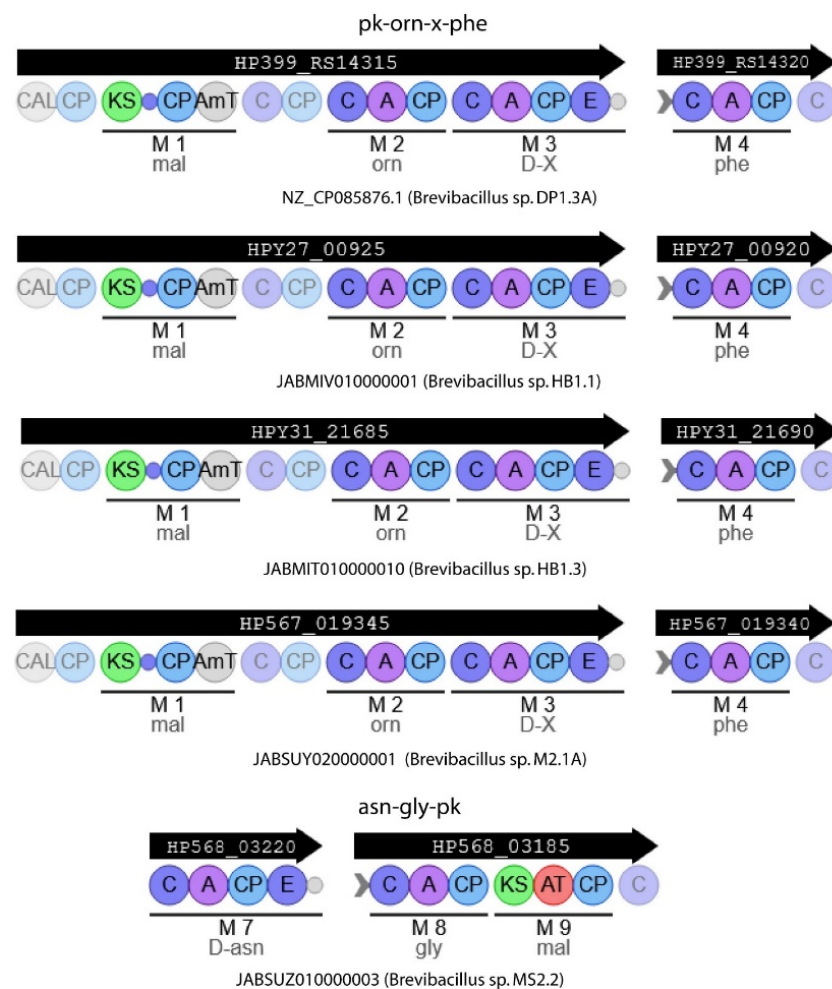


Sactipeptide

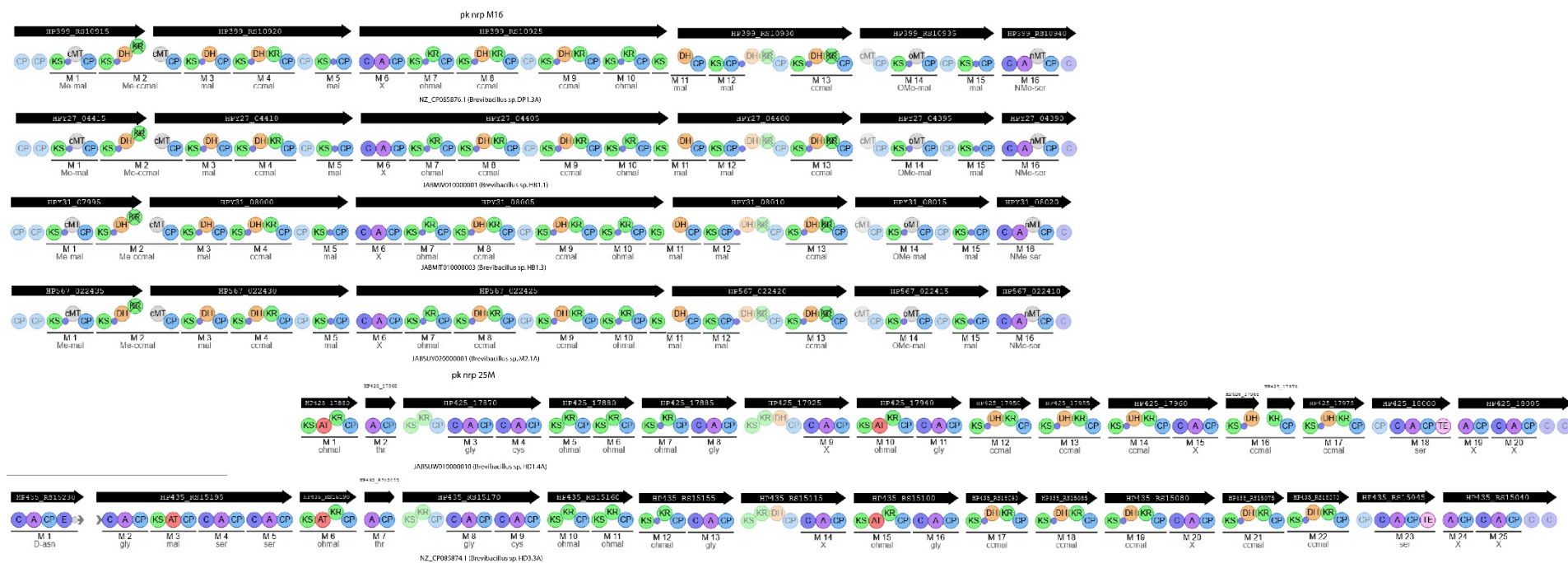
JABSUW010000001.1Brevibacilluspp.HD1.4A1.fasta AOI_03



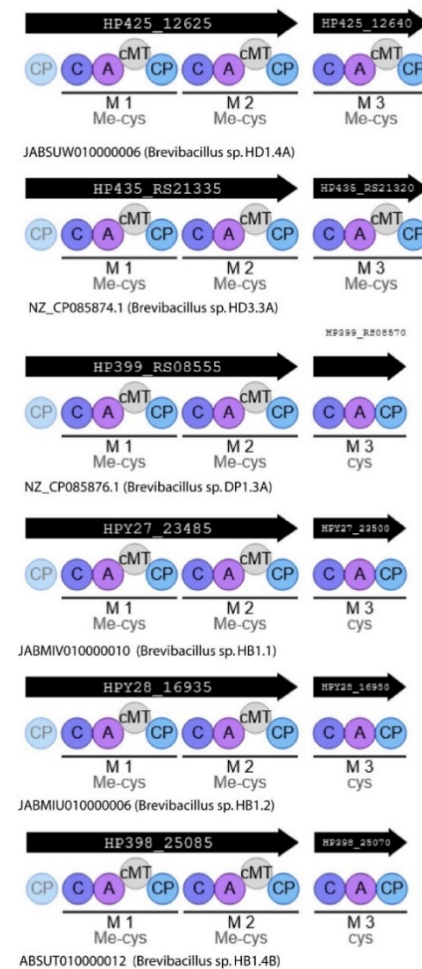
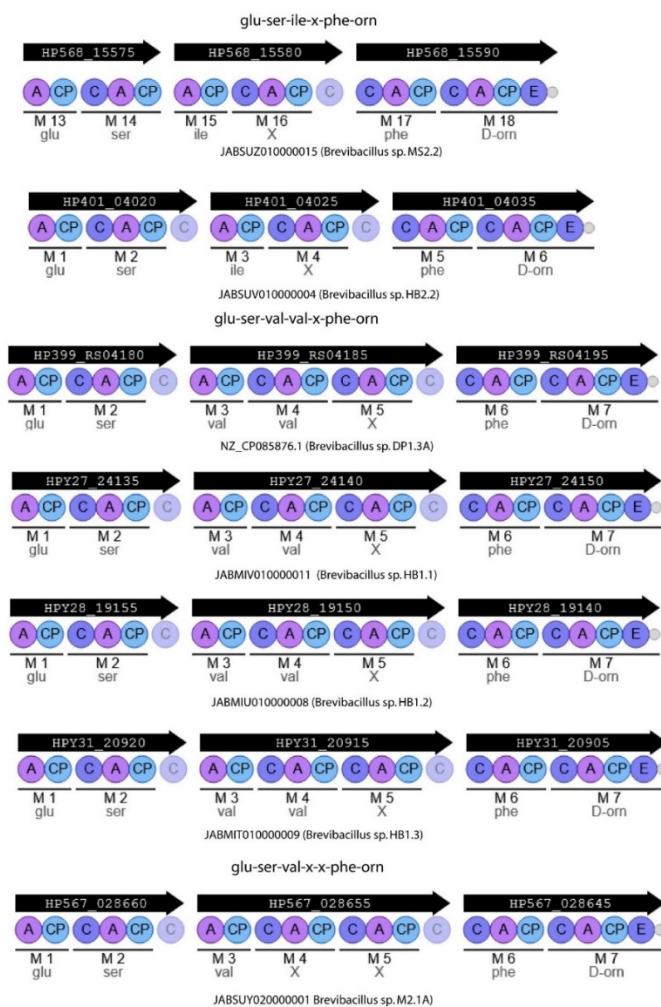
Supplemental Figure S22: UviB and sactipeptides were detected in Brevibacillus HD1.4A by BAGEL4



Supplemental Figure S23: Unknown PK-NRP hybrids pk-orn-x-phe and asn-gly-pk



Supplemental Figure S24: Unknown PK-NRP hybrids: 16M PK-NRP and 20/25M PK-NRP



Left: **Supplemental Figure S25:** Unknown NRPS-like: glu-ser-ile-x-phe-orn/ glu-ser-val-val-x-phe-orn/ glu-ser-val-X-X-phe-D-orn (Brevipentin)

Right: **Supplemental Figure S26:** Unknown NRPS-like: cys-cys-cys (Brevitriazol)