

Supplementary Materials

Genome-Based Classification of *Pedobacter albus* sp. nov. and *Pedobacter flavus* sp. nov. Isolated from Soil

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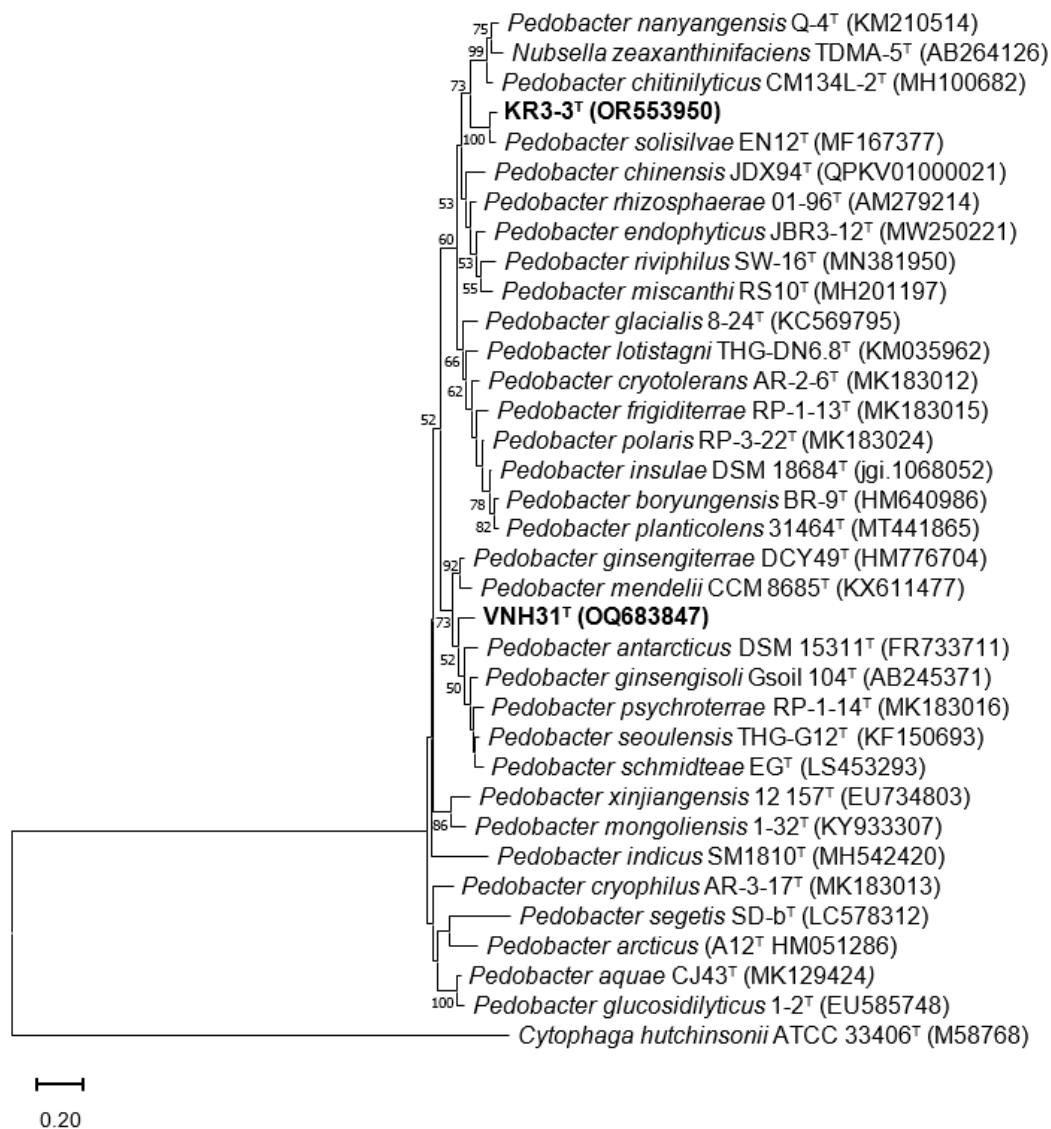


Figure S1. The phylogenetic tree was reconstructed with the maximum likelihood method based on 16S rRNA gene sequences of strain KR3-3^T, VNH31^T, and type strains of the genus *Pedobacter*. *Cytophaga hutchinsonii* ATCC 33406^T was used as an outgroup. Numbers at branch nodes represent confidence levels (values > 50% are shown) from 1000 replicate bootstrap samplings. GenBank accession numbers are shown in parentheses. Bar, 0.20 substitutions per nucleotide position.

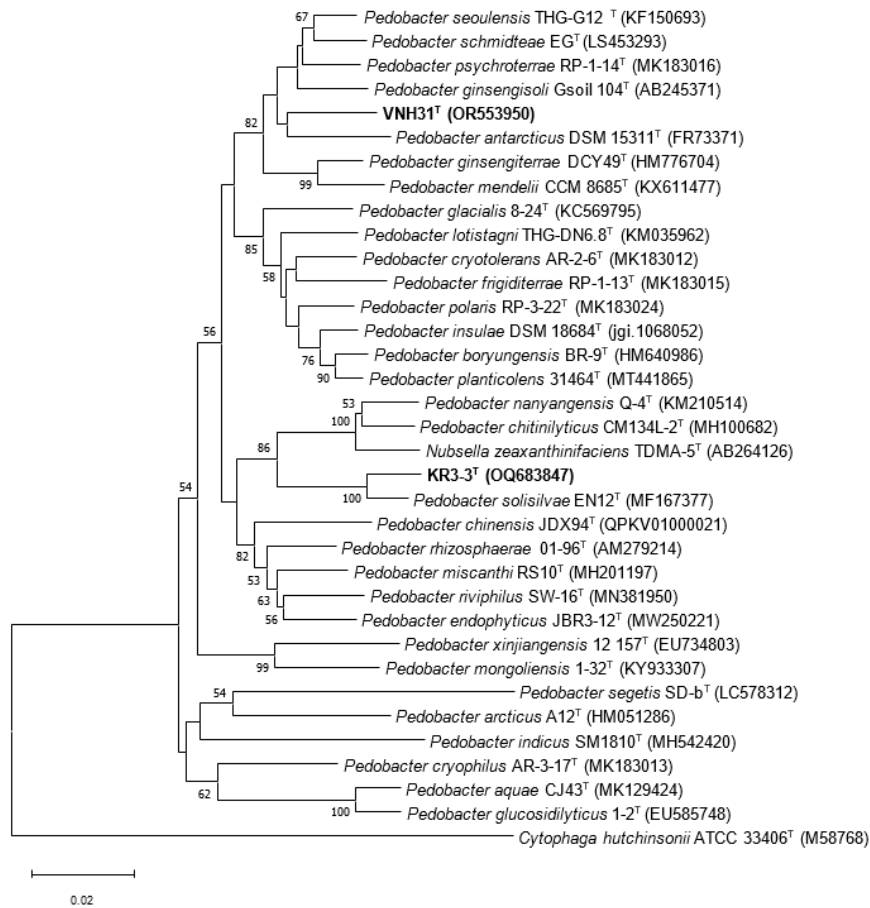


Figure S2. The phylogenetic tree was reconstructed with the minimum evolution method based on 16S rRNA gene sequences of strain KR3-3^T, VNH31^T, and type strains of the genus *Pedobacter*. *Cytophaga hutchinsonii* ATCC 33406^T was used as an outgroup. Numbers at branch nodes represent confidence levels (values > 50% are shown) from 1000 replicate bootstrap samplings. GenBank accession numbers are shown in parentheses. Bar, 0.02 substitutions per nucleotide position.

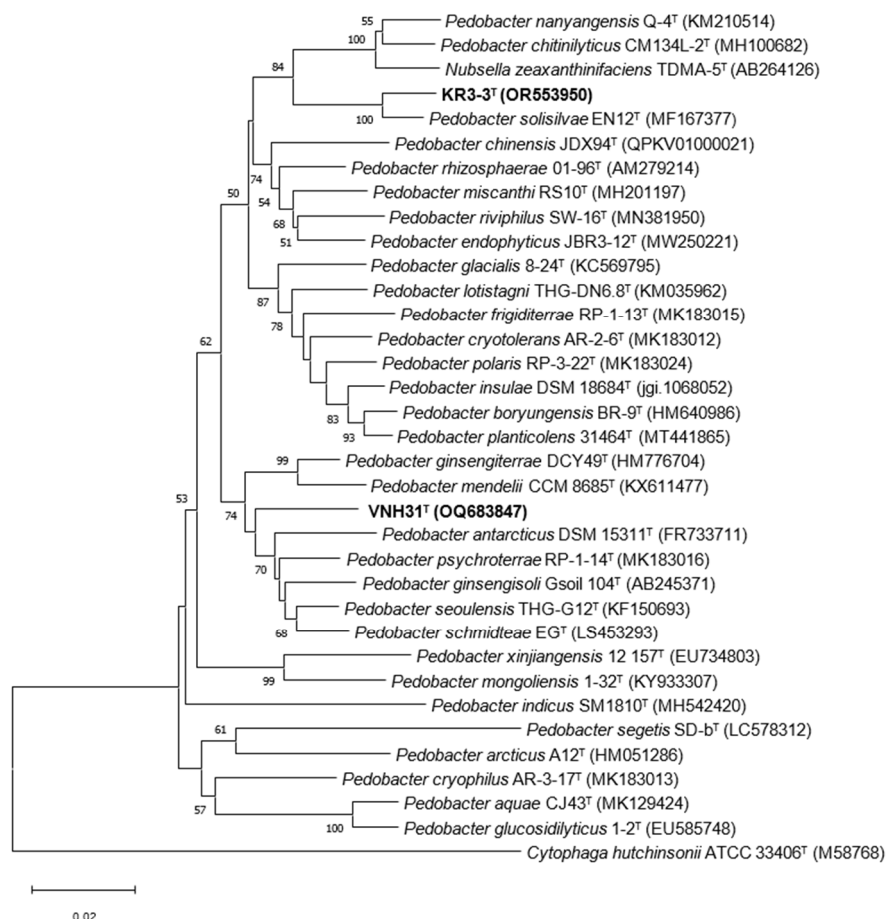


Figure S3. The phylogenetic tree was reconstructed with the neighbor-joining method based on 16S rRNA gene sequences of strain KR3-3, VNH31, and type strains of the genus *Pedobacter*. *Cytophaga hutchinsonii* ATCC 33406^T was used as an outgroup. Numbers at branch nodes represent confidence levels (values > 50 % are shown) from 1000 replicate bootstrap samplings. GenBank accession numbers are shown in parentheses. Bar, 0.02 substitutions per nucleotide position.

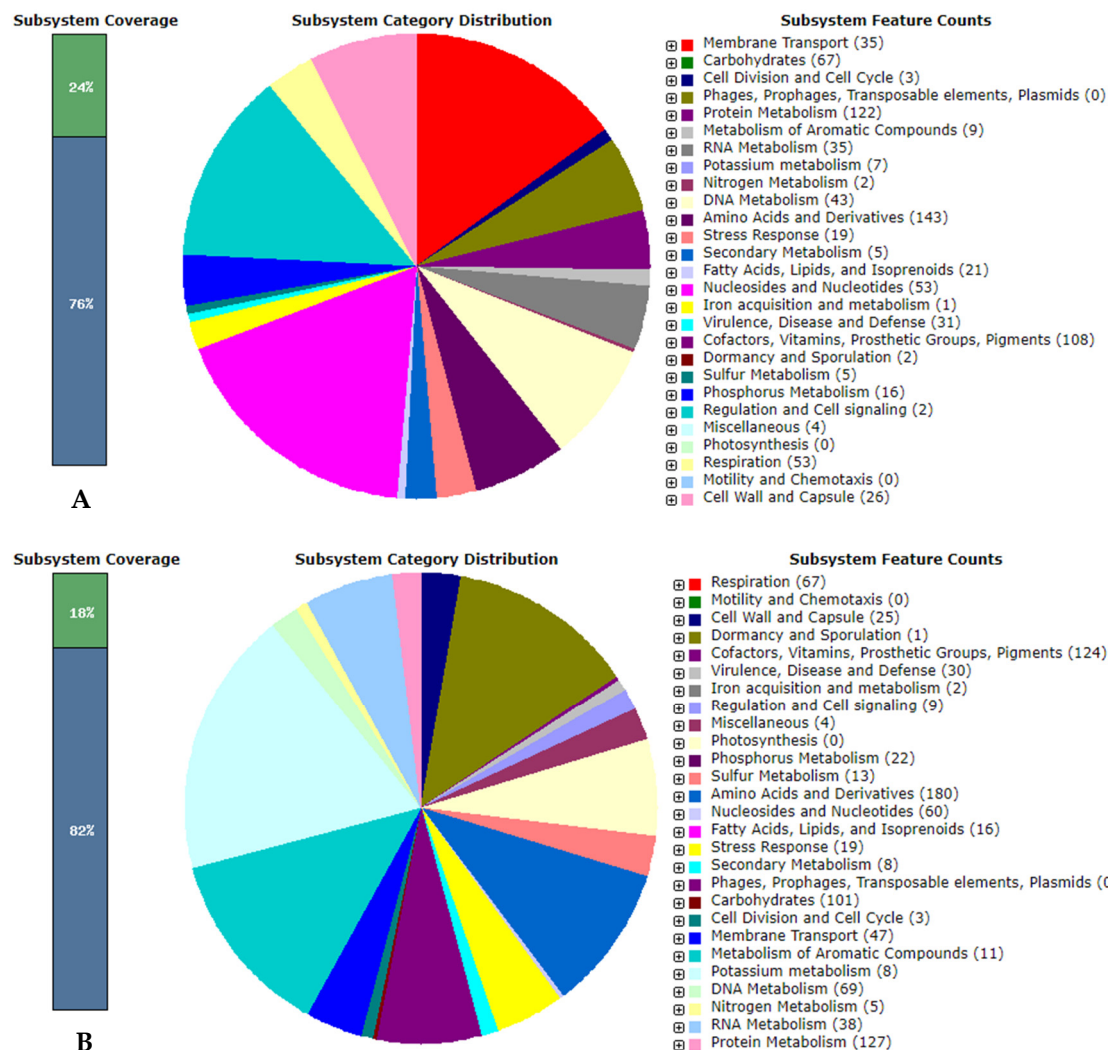


Figure S4. Subsystem feature of strain VNH31^T (A) and KR3-3^T (B) revealed by RAST (Rapid Annotation using Subsystem Technology) server version 2.0.

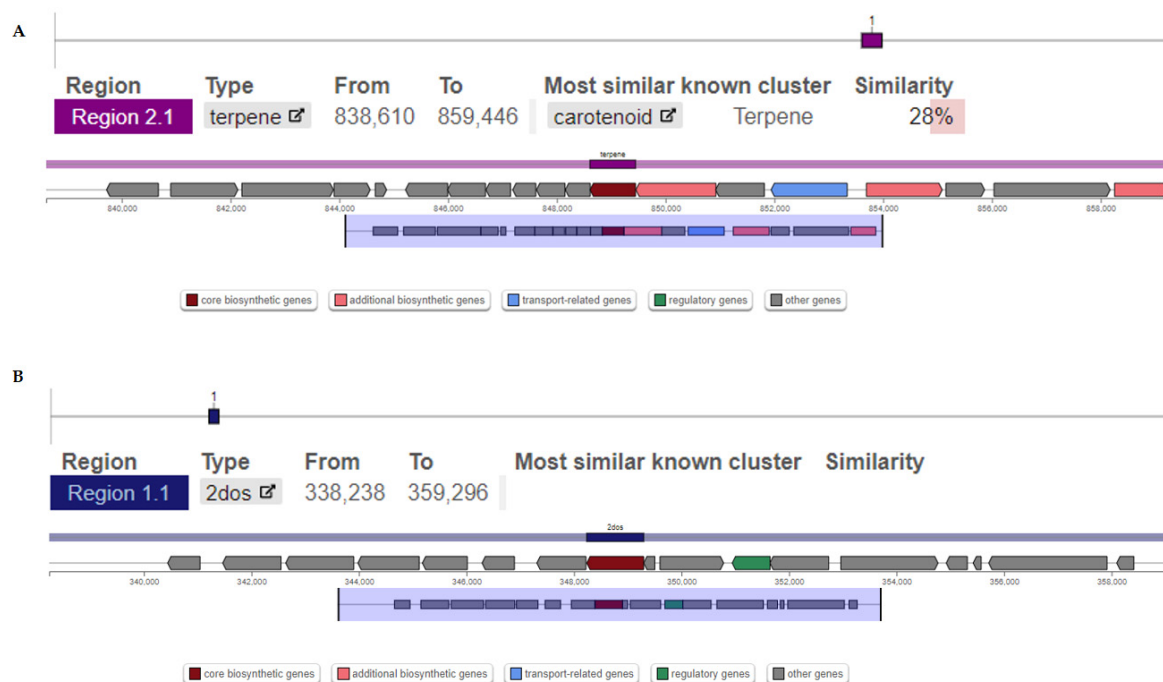


Figure S5. Secondary metabolism analysis in the genome of (A) VNH31^T and (B) KR3-3^T using antiSMASH version 7.1 to predict biosynthetic gene clusters.

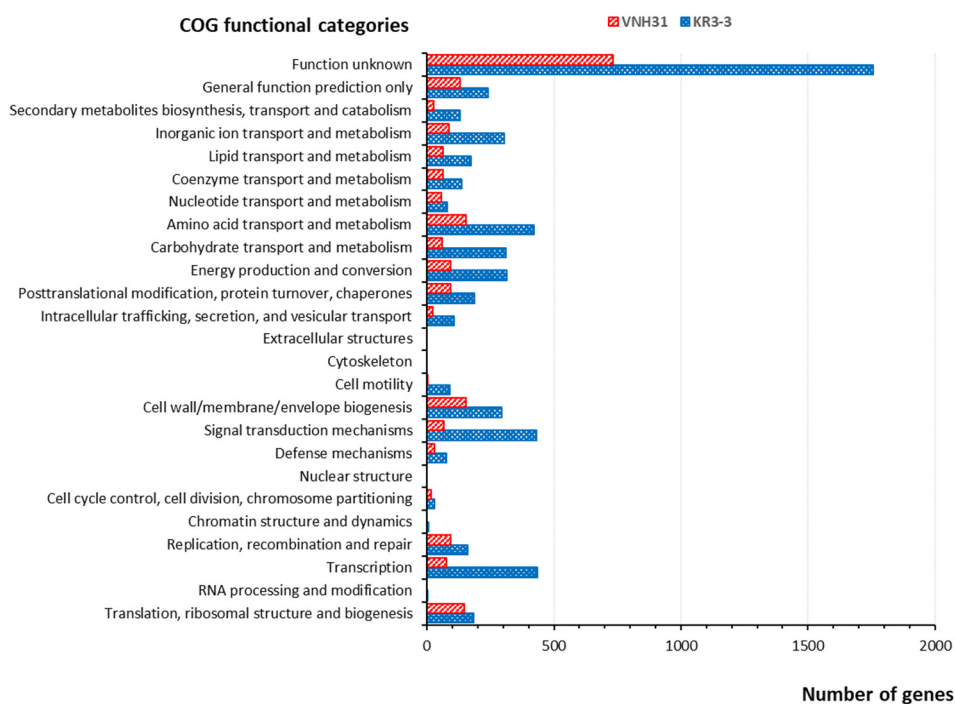


Figure S6. COG functional classification of proteins in strains KR3-3^T and VNH31^T genome.

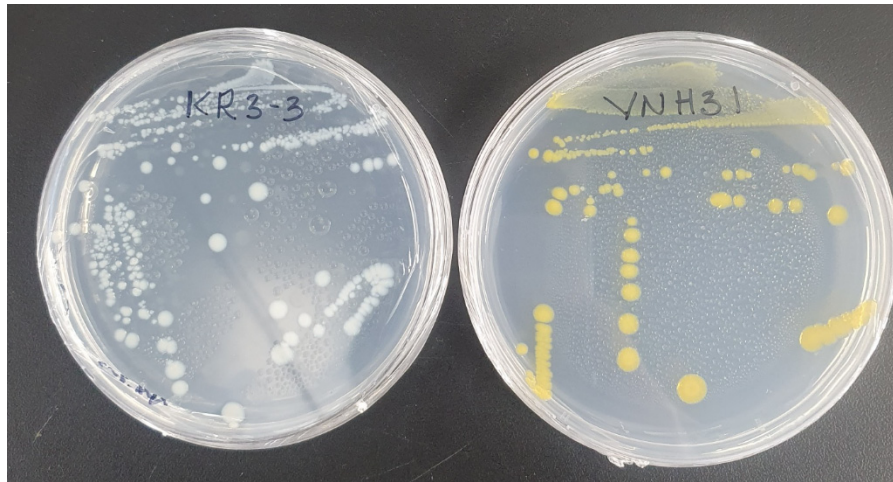


Figure S7. Colonies of KR3-3^T and VNH31^T were grown on R2A at 30°C for 72h.

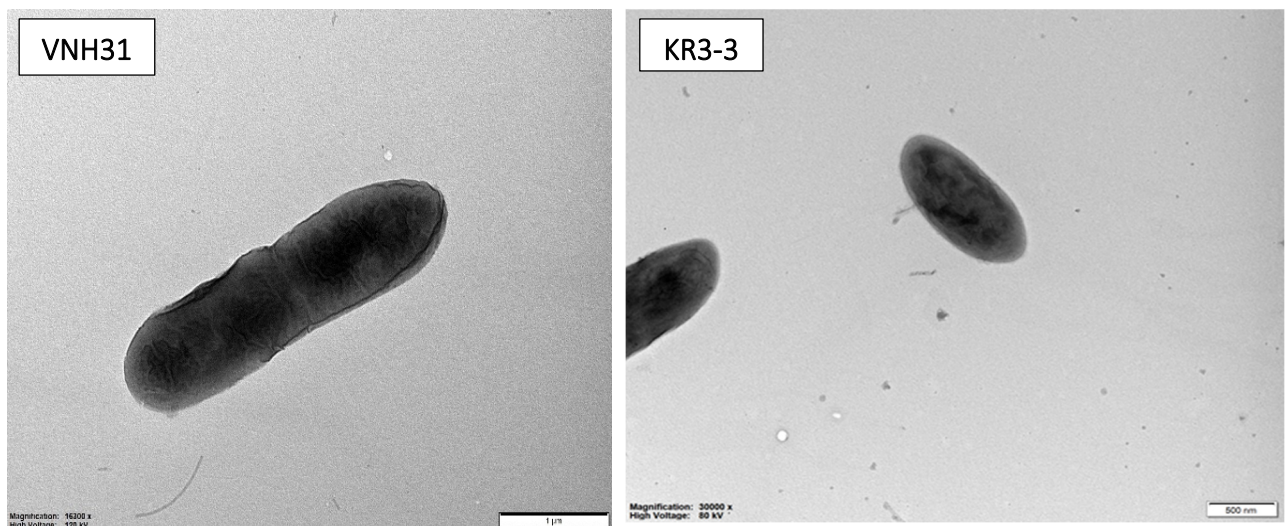


Figure S8. Transmission electron microscopy of strains VNH31^T and KR3-3^T growth on R2A medium plates for 3 days at 30°C. Bar 1 μm with VNH31^T and 500 nm with KR3-3^T

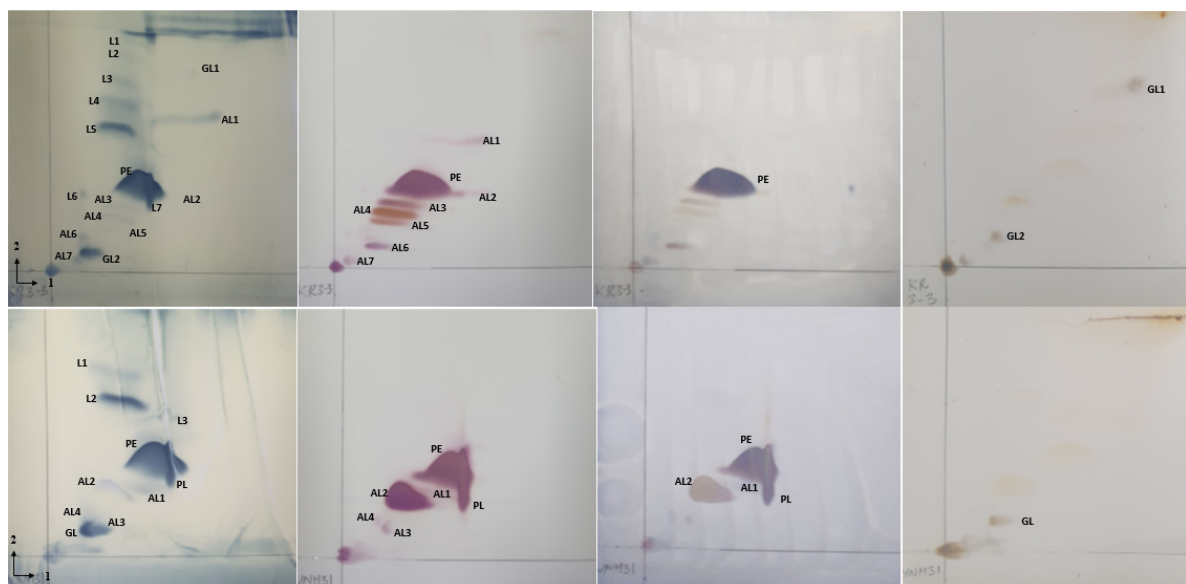


Figure S9. Polar lipid profile of strain KR3-3^T and VNH31^T

PE – phosphatidylethanolamine, GL - unidentified glycolipids, AL – unidentified aminolipid, L – unidentified polar lipids, PL - unidentified phospholipid

Table S1. ANI_b and dDDH values (%) between the genome sequences of the two isolates and other *Pedobacter* species with validly published names.

| No | Taxon name | Strain name | Genome accession no. | KR3-3 ^T | | VNH31 ^T | |
|----|---------------------------|-------------------------|----------------------|--------------------|------------------|--------------------|------------------|
| | | | | dDDH | ANI _b | dDDH | ANI _b |
| 1 | KR3-3 | KR3-3 ^T | JAZDQT000000000.1 | | | 20.00 | 69.86 |
| 2 | VNH31 | VNH31 ^T | JAZDQU000000000.1 | 20.00 | 69.86 | | |
| 3 | <i>P. africanus</i> | DSM 12126 ^T | FWXT01000009.1 | 18.90 | 71.10 | 19.30 | 69.79 |
| 4 | <i>P. agri</i> | PB92 ^T | AJLG01000001.1 | 19.30 | 70.99 | 19.90 | 70.85 |
| 5 | <i>P. alluvionis</i> | DSM 19624 ^T | RCCK01000010.1 | 19.20 | 71.54 | 20.50 | 70.97 |
| 6 | <i>P. antarcticus</i> | ATCC 51969 ^T | GCA_900112985.1 | 19.70 | 70.24 | 20.30 | 70.22 |
| 7 | <i>P. aquae</i> | CJ43 ^T | CP043329.1 | 20.10 | 68.70 | 19.90 | 70.16 |
| 8 | <i>P. aquatilis</i> | CECT 7114 ^T | AUFPW010000001.1 | 18.90 | 70.95 | 20.10 | 71.21 |
| 9 | <i>P. arcticus</i> | A12 ^T | JH947125.1 | 18.90 | 68.56 | 19.20 | 69.46 |
| 10 | <i>P. borealis</i> | DSM 19626 ^T | JAUG01000001.1 | 19.30 | 71.70 | 20.00 | 70.92 |
| 11 | <i>P. boryungensis</i> | LMG 31300 ^T | JABMKV010000001.1 | 19.00 | 73.79 | 19.50 | 71.67 |
| 12 | <i>P. caeni</i> | DSM 16990 ^T | FQUQ01000022.1 | 18.00 | 70.66 | 18.80 | 70.61 |
| 13 | <i>P. changchengzhani</i> | E01020 ^T | SJCY01000001.1 | 19.80 | 70.97 | 19.50 | 71.21 |
| 14 | <i>P. chinensis</i> | JDX94 ^T | QPKV01000013.1 | 18.40 | 71.08 | 22.60 | 71.63 |
| 15 | <i>P. chitinilyticus</i> | CM134L-2 ^T | QMHN01000009.1 | 19.30 | 72.31 | 20.40 | 71.01 |
| 16 | <i>P. cryoconitis</i> | DSM 14825 ^T | QLLR01000083.1 | 20.50 | 70.81 | 18.60 | 70.56 |
| 17 | <i>P. cryophilus</i> | AR-3-17 ^T | SWBP01000001.1 | 20.90 | 68.65 | 19.40 | 70.19 |
| 18 | <i>P. cryotolerans</i> | AR-2-6 ^T | SWBO01000001.1 | 19.20 | 73.37 | 20.20 | 71.83 |
| 19 | <i>P. duraquae</i> | DSM 19034 ^T | SNWM01000001.1 | 19.90 | 70.60 | 19.90 | 69.7 |
| 20 | <i>P. endophyticus</i> | JBR3-12 ^T | CP064939.1 | 19.30 | 71.38 | 20.60 | 70.78 |

| No | Taxon name | Strain name | Genome accession no. | KR3-3 ^T | | VNH31 ^T | |
|----|-----------------------------|---------------------------------|----------------------|--------------------|----------|--------------------|----------|
| | | | | dDD H | ANI b | dDD H | ANI b |
| 21 | <i>P. fastidiosus</i> | CCM 8938 ^T | JACRYL010000010.1 | 18.90 | 71.15 | 20.60 | 71.06 |
| 22 | <i>P. foliorum</i> | LMG 31463 ^T | JABSNU010000010.1 | 19.30 | 71.06 | 18.70 | 71.13 |
| 23 | <i>P. frigidisoli</i> | RP-3-11 ^T | SJSN01000001.1 | 18.90 | 70.92 | 19.50 | 70.99 |
| 24 | <i>P. frigiditerrae</i> | RP-1-13 ^T | SJSK01000001.1 | 18.70 | 73.26 | 19.60 | 71.51 |
| 25 | <i>P. frigoris</i> | RP-3-15 ^T | SWBQ01000001.1 | 19.40 | 70.85 | 18.80 | 70.47 |
| 26 | <i>P. gandavensis</i> | LMG 31462 ^T | WNXC01000010.1 | 18.90 | 70.91 | 20.30 | 70.84 |
| 27 | <i>P. ghigonis</i> | Marseille-Q2390 ^T | CAESCM010000001.1 | 18.90 | 71.70 | 20.00 | 70.61 |
| 28 | <i>P. ginsengisoli</i> | Gsoil 104 ^T | JASSYX010000001.1 | 19.40 | 70.91 | 20.80 | 69.68 |
| 29 | <i>P. ginsenosidimutans</i> | KACC 14530 ^T | LMZQ01000001.1 | 19.10 | 71.52 | 20.50 | 70.89 |
| 30 | <i>P. glucosidilyticus</i> | DSM 23534 ^T | KE384311.1 | 19.50 | 68.84 | 19.30 | 69.97 |
| 31 | <i>P. hartonius</i> | DSM 19033 ^T | FNRA01000040.1 | 19.10 | 70.78 | 19.30 | 69.76 |
| 32 | <i>P. helvus</i> | P-25 ^T | SRMP01000001.1 | 19.80 | 72.05 | 20.90 | 71.15 |
| 33 | <i>P. heparinus</i> | DSM 2366 ^T | CP001681.1 | 19.10 | 71.75 | 19.60 | 69.97 |
| 34 | <i>P. hiemivivus</i> | RP-1-16 ^T | SWDX01000001.1 | 19.10 | 71.16 | 20.10 | 71.06 |
| 35 | <i>P. humicola</i> | GW460-11-11-14-LB5 ^T | CP021237.1 | 19.60 | 71.55 | 20.90 | 70.87 |
| 36 | <i>P. indicus</i> | SM1810 ^T | QRGB01000001.1 | 21.00 | 67.19 | 23.60 | 68.02 |
| 37 | <i>P. insulae</i> | DSM 18684 ^T | FOPP01000020.1 | 19.40 | 72.20 | 20.60 | 70.81 |
| 38 | <i>P. jejuensis</i> | TNB23 ^T | RBEE01000001.1 | 19.10 | 71.07 | 19.50 | 71.04 |
| 39 | <i>P. kyongggii</i> | K-4-11-1 ^T | SIXF01000001.1 | 18.80 | 71.56 | 20.70 | 70.93 |
| 40 | <i>P. kyungheensis</i> | KACC_16221 ^T | JSYN01000001.1 | 19.10 | 71.71 | 19.60 | 70.63 |
| 41 | <i>P. lusitanus</i> | NL19 ^T | JXRA01000001.1 | 19.10 | 70.65 | 18.80 | 70.34 |
| 42 | <i>P. mendelii</i> | CCM 8939 ^T | BMDJ01000001.1 | 19.60 | 71.15 | 19.80 | 71.09 |
| 43 | <i>P. metabolipauper</i> | DSM 19035 ^T | SNYC01000003.1 | 19.40 | 71.18 | 19.10 | 70.29 |
| 44 | <i>P. miscanthi</i> | RS10 ^T | QNQU01000001.1 | 18.60 | 71.78 | 19.40 | 70.51 |
| 45 | <i>P. mongoliensis</i> | KCTC 52859 ^T | JAFMZO010000001.1 | 19.10 | 68.55 | 20.50 | 68.33 |
| 46 | <i>P. montanisoli</i> | CYS-01 ^T | JALGBH010000001.1 | 19.00 | 72.10 | 18.80 | 70.91 |
| 47 | <i>P. mucosus</i> | Q8-18 ^T | CP087585.1 | 20.00 | 70.73 | 21.10 | 71.39 |
| 48 | <i>P. nanyangensis</i> | Q-4 ^T | QMHO01000001.1 | 19.60 | 72.66 | 21.50 | 70.58 |
| 49 | <i>P. nototheniae</i> | 36B243 ^T | GCA_004335085.1 | 18.40 | 70.93 | 19.30 | 71.35 |
| 50 | <i>P. nutrimenti</i> | DSM 27372 ^T | QKLU01000001.1 | 19.30 | 70.93 | 19.40 | 70.03 |
| 51 | <i>P. nyackensis</i> | DSM 19625 ^T | GCA_900176505.1 | 19.20 | 71.26 | 20.10 | 70.75 |
| 52 | <i>P. panaciterrae</i> | LMG 23400 ^T | GCA_013204795.1 | 18.70 | 70.83 | 19.20 | 70.82 |
| 53 | <i>P. petrophilus</i> | LMG 29686 ^T | WKKH01000001.1 | 19.20 | 70.94 | 19.70 | 70.74 |
| 54 | <i>P. planticolens</i> | LMG 31464 ^T | WNXD01000001.1 | 19.20 | 74.19 | 18.80 | 71.48 |
| 55 | <i>P. polaris</i> | RP-3-22 ^T | SWBR01000001.1 | 18.70 | 73.29 | 19.70 | 71.66 |
| 56 | <i>P. psychrodurus</i> | RP-3-21 ^T | SJSO01000001.1 | 19.20 | 71.53 | 20.10 | 70.28 |
| 57 | <i>P. psychrophilus</i> | CCM 8644 ^T | LWHJ01000001.1 | 19.70 | 68.55 | 19.20 | 69.96 |
| 58 | <i>P. psychroterrae</i> | RP-1-14 ^T | SJSL01000001.1 | 18.80 | 70.99 | 19.40 | 69.91 |
| 59 | <i>P. psychrotolerans</i> | DSM 103236 ^T | SLWO01000001.1 | 19.00 | 70.85 | 20.00 | 70.73 |
| 60 | <i>P. quisquiliarum</i> | CGMCC | BMIL01000001.1 | 20.30 | 70.44 | 19.20 | 69.74 |

| No | Taxon name | Strain name | Genome accession no. | KR3-3 ^T | | VNH31 ^T | |
|----|--------------------------|---------------------------------|----------------------|--------------------|----------|--------------------|----------|
| | | | | dDD H | ANI b | dDD H | ANI b |
| | | 1.15343 ^T | | | | | |
| 61 | <i>P. rhizosphaerae</i> | DSM 18610 ^T | GCA_900111155.1 | 18.90 | 70.90 | 20.50 | 70.61 |
| 62 | <i>P. riviphilus</i> | SW-16 ^T | CP061171.1 | 19.70 | 71.39 | 20.60 | 70.88 |
| 63 | <i>P. roseus</i> | KACC 11594 ^T | CP060723.1 | 19.70 | 71.45 | 20.30 | 70.77 |
| 64 | <i>P. sandarakinus</i> | AMB ^T | JAPIVG010000001.1 | 18.80 | 70.59 | 19.50 | 70.46 |
| 65 | <i>P. schmidteae</i> | EG ^T | GCA_900564155.1 | 19.80 | 71.57 | 21.50 | 70.32 |
| 66 | <i>P. segetis</i> | SD-b ^T | JAEHFY010000010.1 | 19.90 | 68.75 | 19.70 | 69.61 |
| 67 | <i>P. soli</i> | DSM 18609 ^T | GCA_900101435.1 | 19.30 | 71.67 | 19.00 | 70.58 |
| 68 | <i>P. steynii</i> | DSM 19110 ^T | FNGY01000041.1 | 18.70 | 70.79 | 19.70 | 70.47 |
| 69 | <i>P. suwonensis</i> | Fj 001 ^T | CP031708.1 | 19.90 | 71.44 | 20.90 | 70.64 |
| 70 | <i>P. terrae</i> | DSM 17933 ^T | FNCH01000063.1 | 19.20 | 71.62 | 20.00 | 70.83 |
| 71 | <i>P. ureilyticus</i> | THG-T11 ^T | SSHJ01000001.1 | 19.40 | 72.36 | 20.80 | 71.24 |
| 72 | <i>P. westerhofensis</i> | DSM 19036 ^T | FXTN01000001.1 | 18.40 | 70.55 | 18.80 | 69.87 |
| 73 | <i>P. xinjiangensis</i> | CCTCC AB 208092 ^T | JAFMZP010000010.1 | 19.40 | 67.86 | 21.40 | 68.34 |
| 74 | <i>P. xixisoli</i> | CGMCC 1.12803 ^T | OCMT01000007.1 | 19.90 | 72.14 | 20.40 | 71.29 |
| 75 | <i>P. yulinensis</i> | YL28-9 ^T | KZ686268.1 | 18.30 | 69.79 | 17.60 | 67.67 |

Table S2. Results from API ZYM, API 20NE, API 32GN test. 1, VNH31^T; 2, KR3-3^T; 3, *Pedobacter solisilvae* KCTC 42612^T; 4, *Pedobacter insulae* KACC 14010^T; 5, *Pedobacter cryotolerans* KACC 19998^T(data are from the present study, except G+C content of reference strains, which were retrieved from the literature in parentheses); 6, *Pedobacter cryophilus* KACC 19999^T; 7, *Pedobacter mongoliensis* KCTC 52859^T. +, positive; -, negative; +/-, weak positive or ambiguous; nd, no data.

| Characteristics | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|-------------------------------------|---|---|---|-----|---|-----|---|
| Enzymatic reaction (API ZYM) | | | | | | | |
| Alkaline phosphate | + | + | + | - | + | + | + |
| Esterase (C4) | + | + | + | - | - | +/- | + |
| Esterase lipase (C8) | + | + | + | +/- | + | +/- | + |
| Lipase (C14) | - | - | - | - | - | - | - |
| Leucine arylamidase | + | + | + | + | + | + | + |
| Valine arylamidase | + | + | + | + | + | + | - |
| Crystine arylamidase | + | + | + | - | + | + | - |
| Trypsin | - | - | + | - | + | - | - |
| Alpha chymotrypsin | + | - | + | + | + | - | - |
| Acid phosphatase | + | + | + | + | + | + | + |
| Naphthol-AS-BI- phosphate | + | + | + | + | + | + | + |
| Alpha galactosidase | - | + | + | - | - | - | - |
| Beta galactosidase | - | + | + | - | - | + | + |
| Beta glucuronidase | - | - | - | - | - | - | - |
| Alpha glucosidase | + | + | + | + | + | - | - |
| Beta glucosidase | - | + | + | + | + | - | + |
| N-acetyl beta | + | + | + | + | + | + | + |

| Characteristics | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---------------------------|-----|-----|---|---|---|-----|---|
| glucosaminidase | | | | | | | |
| Alpha monosidase | - | - | - | - | - | - | - |
| Alpha fucosidase | - | + | + | - | - | - | - |
| API 20NE | | | | | | | |
| Nitrate reduction | - | - | - | - | - | - | - |
| Indole production | - | - | - | - | - | - | - |
| Glucose fermentation | - | - | - | - | - | - | - |
| Arginine dihydrolase | - | - | - | - | - | | - |
| Urease | - | - | - | - | - | - | - |
| Aesculin hydrolysis | - | + | + | + | + | + | + |
| Gelatin hydrolysis | - | - | - | - | - | - | - |
| β - Galactosidase | + | + | + | - | - | + | + |
| Assimilation of | | | | | | | |
| D-Glucose | +/- | + | + | + | + | + | + |
| L-Arabinose | - | - | - | + | + | + | + |
| D-Mannose | - | + | + | + | + | +/- | + |
| D-Mannitol | - | - | - | - | - | nd | - |
| N-Acetyl-glucosamine | +/- | + | + | - | + | + | + |
| D-Maltose | - | + | + | + | + | nd | - |
| Potassium gluconate | - | - | - | + | - | - | - |
| Capric acid | - | - | - | - | - | nd | - |
| Adipic acid | - | - | - | - | - | nd | - |
| Malate | - | - | - | + | - | - | - |
| Trisodium citrate | - | - | - | - | - | nd | - |
| Phenylacetic acid | - | - | - | - | - | nd | - |
| API 32GN | | | | | | | |
| L-Rhamnose | - | - | - | - | + | + | - |
| N-Acetyl-glucosamine | - | + | + | + | + | nd | + |
| D-Ribose | - | - | - | - | - | - | - |
| Inositol | - | - | - | + | - | nd | - |
| D-Saccharose (sucrose) | - | + | + | + | - | - | - |
| D-Maltose | - | + | + | + | + | - | - |
| Itaconic acid | +/- | - | - | - | - | nd | - |
| Suberic acid | - | - | - | - | - | +/- | - |
| Sodium malonate | - | - | - | - | - | nd | - |
| Sodium acetate | + | + | + | - | - | - | - |
| Lactic acid | - | +/- | - | - | - | - | - |
| L-Alanine | - | - | - | + | - | | - |
| Potassium 5-ketogluconate | - | - | - | - | - | - | - |
| Glycogen | - | - | - | + | + | - | - |
| 3-Hydroxybenzoic acid | - | - | - | - | - | - | - |
| L-Serine | + | + | - | + | - | - | - |
| D-Mannitol | - | - | - | - | - | - | - |
| D-Glucose | - | + | + | + | + | +/- | + |
| Salicin | - | + | + | + | - | - | - |
| D-Melibiose | - | + | + | + | - | - | - |

| Characteristics | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---------------------------|-----|-----|-----|---|---|-----|---|
| L-Fucose | +/- | +/- | - | - | - | - | - |
| D-Sorbitol | - | - | - | - | - | - | - |
| L-Arabinose | +/- | - | - | + | + | +/- | + |
| Propionic acid | - | - | - | - | - | - | - |
| Capric acid | - | - | - | - | - | nd | - |
| Valeric acid | - | - | - | - | - | - | - |
| Trisodium citrate | - | - | - | - | - | nd | - |
| L-Histidine | - | - | - | - | - | +/- | - |
| Potassium 2-ketogluconate | - | - | - | - | - | - | - |
| 3-Hydroxybutyric acid | - | - | +/- | + | - | - | - |
| 4-Hydroxybenzoic acid | - | + | - | - | - | - | - |
| L-Proline | + | + | + | + | - | + | - |