

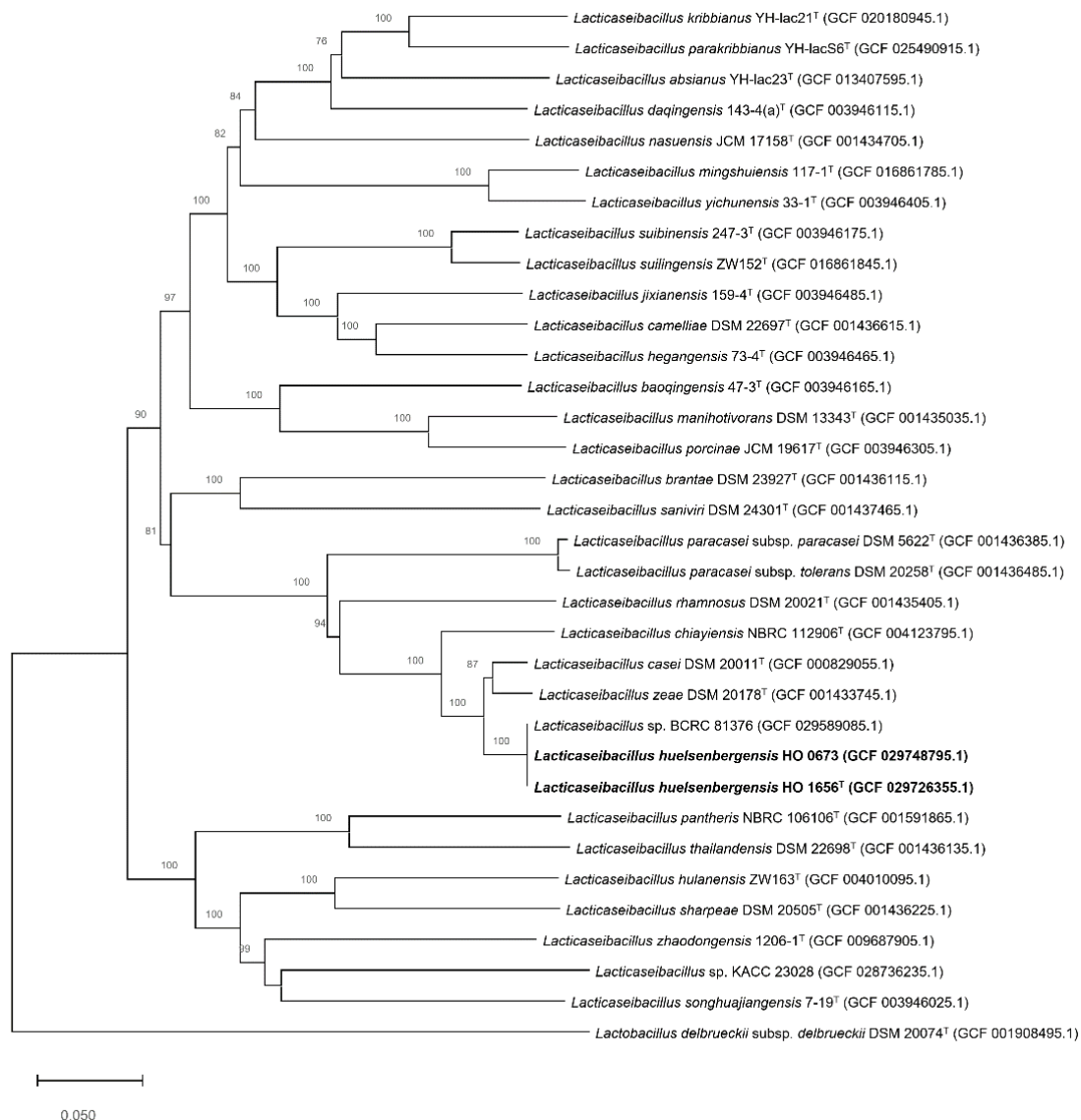
## 1. Supplementary Material

**Supplementary Table 1.** BLAST analysis of 16S rRNA gene and the concatenated *dnaK\_mutL\_pheS* genes of HO 1656<sup>T</sup> and related species.

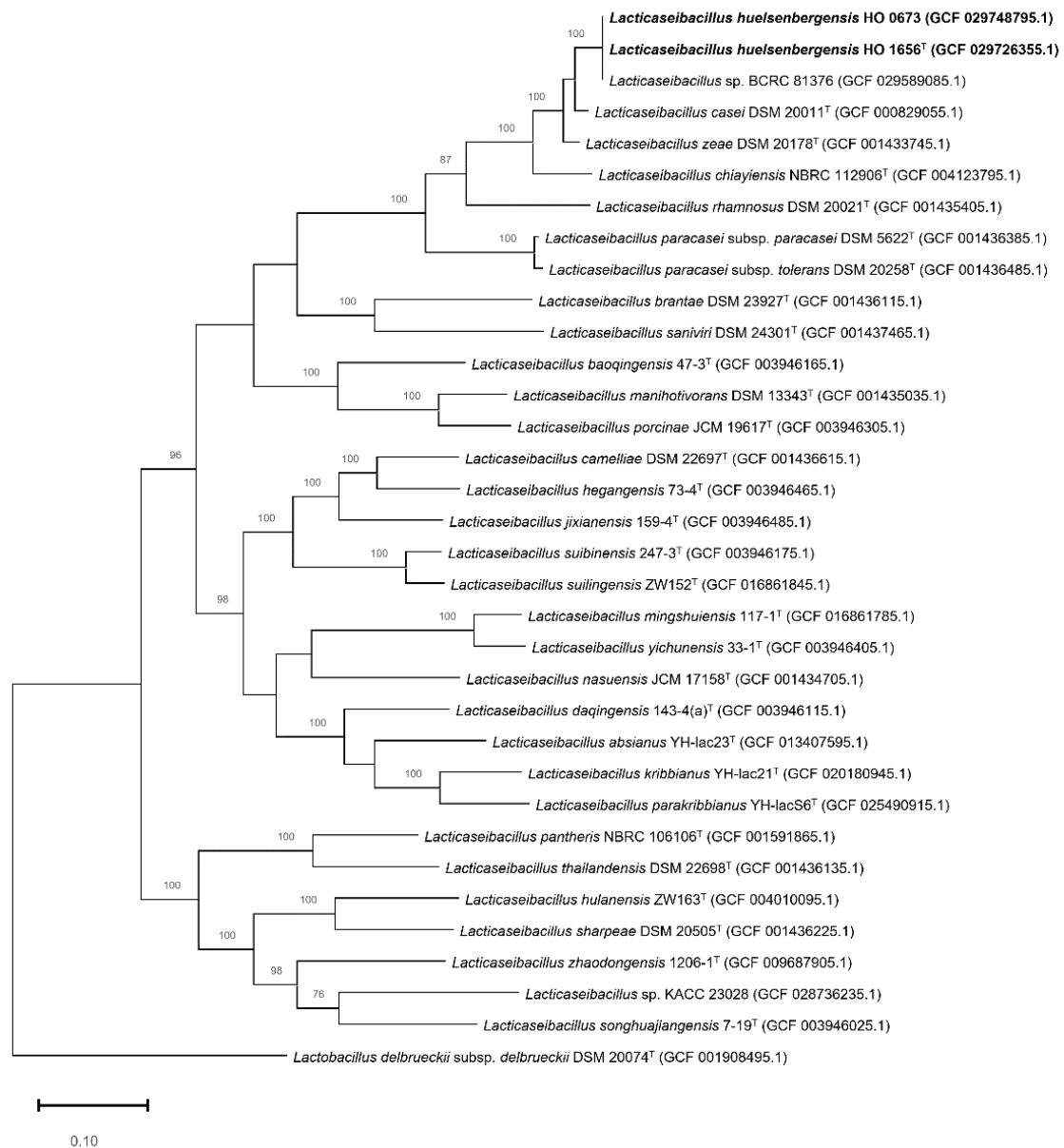
Species (accession number)	16S rRNA identity	<i>dnaK_mutL_pheS</i> identity
HO 1656 <sup>T</sup> (GCF_029726355.1)	100.0	100.0
<i>Lactcaseibacillus</i> sp. BCRC 81376 (GCF_029589085.1)	99.7	100.0
<i>Lactcaseibacillus casei</i> DSM 20011 <sup>T</sup> (GCF_000829055.1)	99.7	96.3
<i>Lactcaseibacillus chiayiensis</i> NBRC 112906 <sup>T</sup> (GCF_004123795.1)	99.7	90.8
HO 0673 (GCF_029748795.1)	99.6	100.0
<i>Lactcaseibacillus zeae</i> DSM 20178 <sup>T</sup> (GCF_001433745.1)	99.5	95.2
<i>Lactcaseibacillus paracasei</i> subsp. <i>tolerans</i> DSM 20258 <sup>T</sup> (GCF_001436485.1)	99.4	84.0
<i>Lactcaseibacillus paracasei</i> subsp. <i>paracasei</i> DSM 5622 <sup>T</sup> (GCF_001436385.1)	99.1	82.0
<i>Lactcaseibacillus rhamnosus</i> DSM 20021 <sup>T</sup> (GCF_001435405.1)	98.9	82.9
<i>Lactcaseibacillus mingshuiensis</i> 117-1 <sup>T</sup> (GCF_016861785.1)	96.5	74.6
<i>Lactcaseibacillus yichunensis</i> 33-1 <sup>T</sup> (GCF_003946405.1)	96.4	74.4
<i>Lactcaseibacillus suilingensis</i> ZW152 <sup>T</sup> (GCF_016861845.1)	95.6	75.7
<i>Lactcaseibacillus baoqingensis</i> 47-3 <sup>T</sup> (GCF_003946165.1)	95.6	74.3
<i>Lactcaseibacillus suibinensis</i> 247-3 <sup>T</sup> (GCF_003946175.1)	95.5	79.1
<i>Lactcaseibacillus brantae</i> DSM 23927 <sup>T</sup> (GCF_001436115.1)	95.3	76.1
<i>Lactcaseibacillus saniviri</i> DSM 24301 <sup>T</sup> (GCF_001437465.1)	95.1	75.4
<i>Lactcaseibacillus jixianensis</i> 159-4 <sup>T</sup> (GCF_003946485.1)	95.1	79.0
<i>Lactcaseibacillus parakribbianus</i> YH-lacS6 <sup>T</sup> (GCF_025490915.1)	95.0	73.8
<i>Lactcaseibacillus hegangensis</i> 73-4 <sup>T</sup> (GCF_003946465.1)	94.9	75.0
<i>Lactcaseibacillus kribbianus</i> YH-lac21 <sup>T</sup> (GCF_020180945.1)	94.9	73.9
<i>Lactcaseibacillus camelliae</i> DSM 22697 <sup>T</sup> (GCF_001436615.1)	94.7	78.9
<i>Lactcaseibacillus daqingensis</i> 143-4(a) <sup>T</sup> (GCF_003946115.1)	94.3	75.0
<i>Lactcaseibacillus manihotivorans</i> DSM 13343 <sup>T</sup> (GCF_001435035.1)	94.3	74.6
<i>Lactcaseibacillus porcinae</i> JCM 19617 <sup>T</sup> (GCF_003946305.1)	94.3	74.5
<i>Lactcaseibacillus absianus</i> YH-lac23 <sup>T</sup> (GCF_013407595.1)	94.2	76.3
<i>Lactcaseibacillus zhaodongensis</i> 1206-1 <sup>T</sup> (GCF_009687905.1)	94.2	72.9
<i>Lactcaseibacillus nasuensis</i> JCM 17158 <sup>T</sup> (GCF_001434705.1)	94.0	79.0
<i>Lactcaseibacillus songhuajiangensis</i> 7-19 <sup>T</sup> (GCF_003946025.1)	93.7	78.7
<i>Lactcaseibacillus thailandensis</i> DSM 22698 <sup>T</sup> (GCF_001436135.1)	93.5	78.6
<i>Lactcaseibacillus pantheris</i> NBRC 106106 <sup>T</sup> (GCF_001591865.1)	93.3	73.5
<i>Lactcaseibacillus</i> sp. KACC 23028 (GCF_028736235.1)	93.3	78.2
<i>Lactcaseibacillus sharpeae</i> DSM 20505 <sup>T</sup> (GCF_001436225.1)	93.2	73.4
<i>Lactcaseibacillus hulanensis</i> ZW163 <sup>T</sup> (GCF_004010095.1)	92.8	77.1

**Supplementary Table 2.** Assembly statistics of the whole genome sequence assembly of HO 1656<sup>T</sup> and HO 0673.

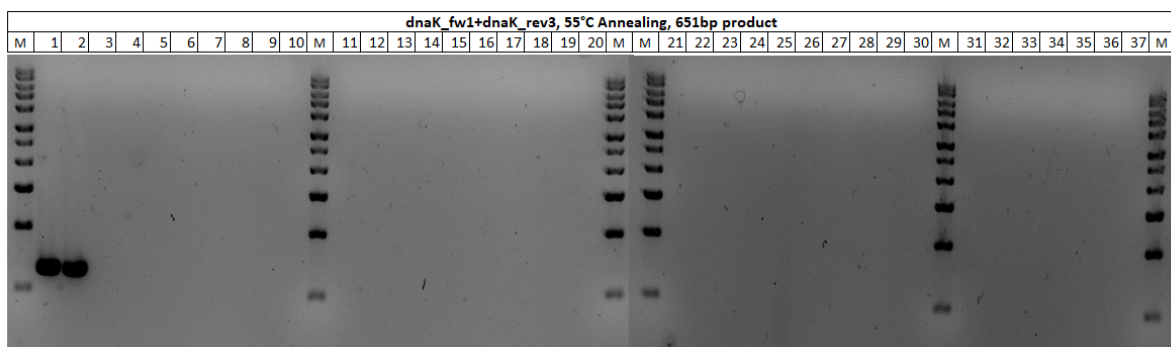
	HO 1656 <sup>T</sup>	HO 0673
accession number	GCA_029726355.1	GCA_029748795.1
genome size	3,094,327 bp	3,134,028 bp
N50	3,094,327 bp	3,134,028 bp
Coverage	807.3	481.4
G and C content [ml%]	48.0	48.0
	1 contig per replicon. Here 1 chromosome, no plasmids.	1 contig per replicon. Here 1 chromosome, no plasmids.
prokka annotation		
number of annotated features		
CDS	2798	2894
total RNA	108	111
ncRNAs	33	36
rRNAs	15	15
t(m)RNAs	60	60



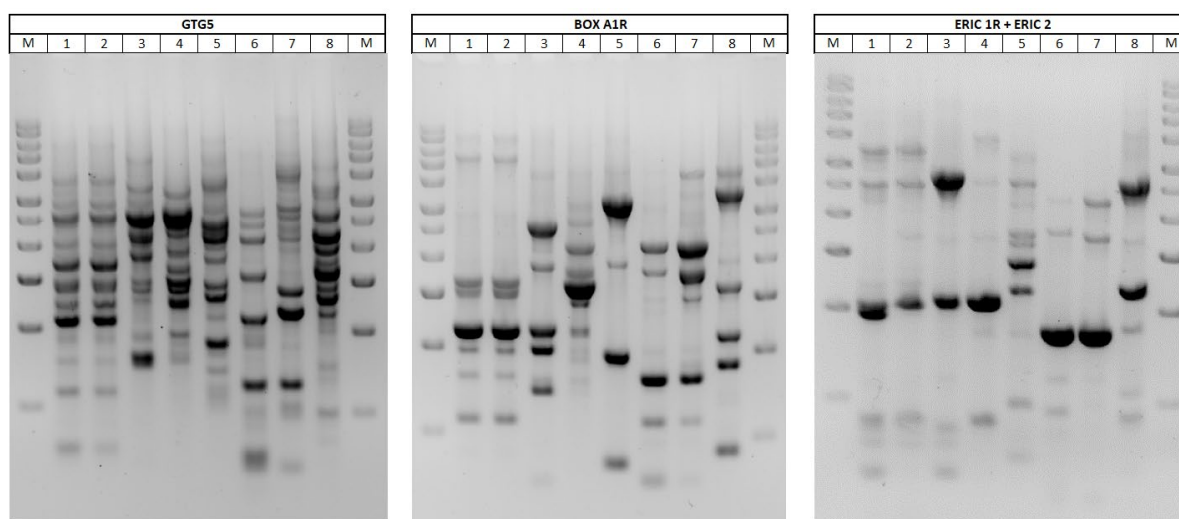
**Supplementary Figure 1.** Phylogenetic tree based on the concatenated sequences of the *dnaK*, *mutL* and *pheS* genes, reconstructed with the neighbour-joining algorithm, indicating the position of the strains HO 1656<sup>T</sup> and HO 0673 in the *Lactocaseibacillus* group. *Lactobacillus delbrueckii* subsp. *delbrueckii* DSM 20074<sup>T</sup> served as outgroup. Bootstrap values greater than 70% based on 1000 replicates are indicated at each node. Bar, 0.05 substitutions per nucleotide position.



**Supplementary Figure 2.** Phylogenetic tree based on the concatenated sequences of the *dnaK*, *mutL* and *pheS* genes, reconstructed with the maximum-likelihood algorithm, indicating the position of the strains HO 1656<sup>T</sup> and HO 0673 in the *Lactocaseibacillus* group. *Lactobacillus delbrueckii* subsp. *delbrueckii* DSM 20074<sup>T</sup> served as outgroup. Bootstrap values greater than 70% based on 1000 replicates are indicated at each node. Bar, 0.10 substitutions per nucleotide position.

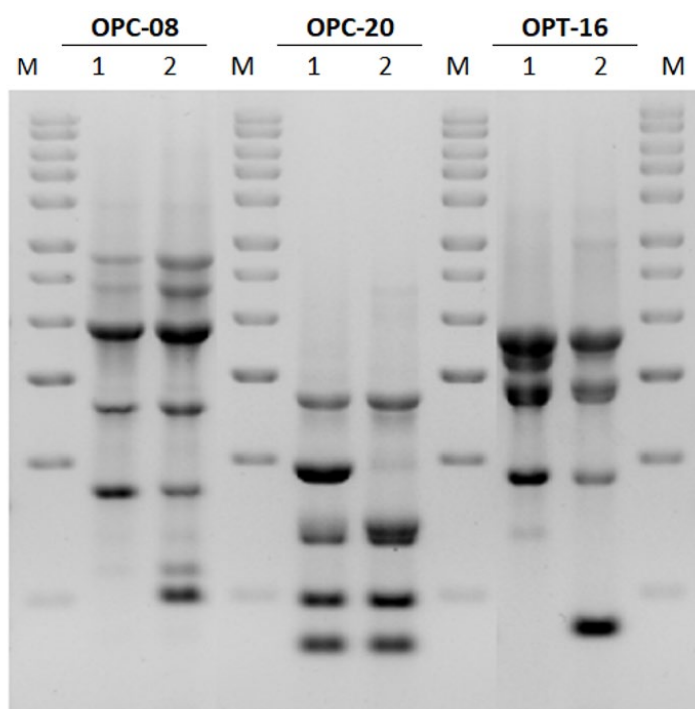


**Supplementary Figure 3.** PCR with species specific primers. M, 1kb DNA marker (Roth); 1, HO 1656<sup>T</sup> (GCF\_029726355.1); 2, HO 0673 (GCF\_029748795.1); 3, *L. casei* DSM 20011<sup>T</sup>; 4, *L. zae* DSM 20178<sup>T</sup>; 5, *L. chiayiensis* NBRC112906<sup>T</sup>; 6, *L. paracasei* subsp. *paracasei* DSM 5622<sup>T</sup>; 7, *L. paracasei* subsp. *tolerans* DSM 20258<sup>T</sup>; 8, *L. rhamnosus* DSM 20021<sup>T</sup>; 9, *S. perolens* DSM 12744<sup>T</sup>; 10, *L. yichunensis* NCIMB 15169<sup>T</sup>; 11, *L. diolivorans* DSM 14221<sup>T</sup>; 12, *L. reuteri* DSM 20016<sup>T</sup>; 13, *L. sakei* subsp. *sakei* DSM 20017<sup>T</sup>; 14, *L. buchneri* subsp. *buchneri* DSM 20057<sup>T</sup>; 15, *L. brevis* DSM 20054<sup>T</sup>; 16, *Lactococcus cremoris* subsp. *cremoris* DSM 20069<sup>T</sup>; 17, *L. lactis* subsp. *lactis* DSM 20481<sup>T</sup>; 18, *L. delbrucknerii* subsp. *bulgaricus* DSM 20081<sup>T</sup>; 19, *L. farraginis* DSM 18382<sup>T</sup>; 20, *L. parafarraginis* DSM 18390<sup>T</sup>; 21, *L. curvatus* DSM 20019<sup>T</sup>; 22, *L. plantarum* subsp. *plantarum* DSM 20174<sup>T</sup>; 23, *F. sanfransiscensis* DSM 20451<sup>T</sup>; 24, *L. amylovorus* DSM 20531<sup>T</sup>; 25, *L. fermentum* DSM 20052<sup>T</sup>; 26, *S. collinoides* DSM 20515<sup>T</sup>; 27, *L. acidophilus* DSM 20079<sup>T</sup>; 28, *L. frumenti* DSM 13145<sup>T</sup>; 29, *L. kefir* DSM 20587<sup>T</sup>; 30, *L. vaginalis* DSM 5837<sup>T</sup>; 31, *L. coryniformis* subsp. *coryniformis* DSM 20001<sup>T</sup>; 32, *L. rapi* DSM 19907<sup>T</sup>; 33, *L. pentosus* DSM 20314<sup>T</sup>; 34, *L. kisonensis* DSM 19906<sup>T</sup>; 35, *L. hilgardii* DSM 20176<sup>T</sup>; 36, *L. parabuchneri* DSM 5707<sup>T</sup>; 37, *L. gasseri* DSM 20243<sup>T</sup>.

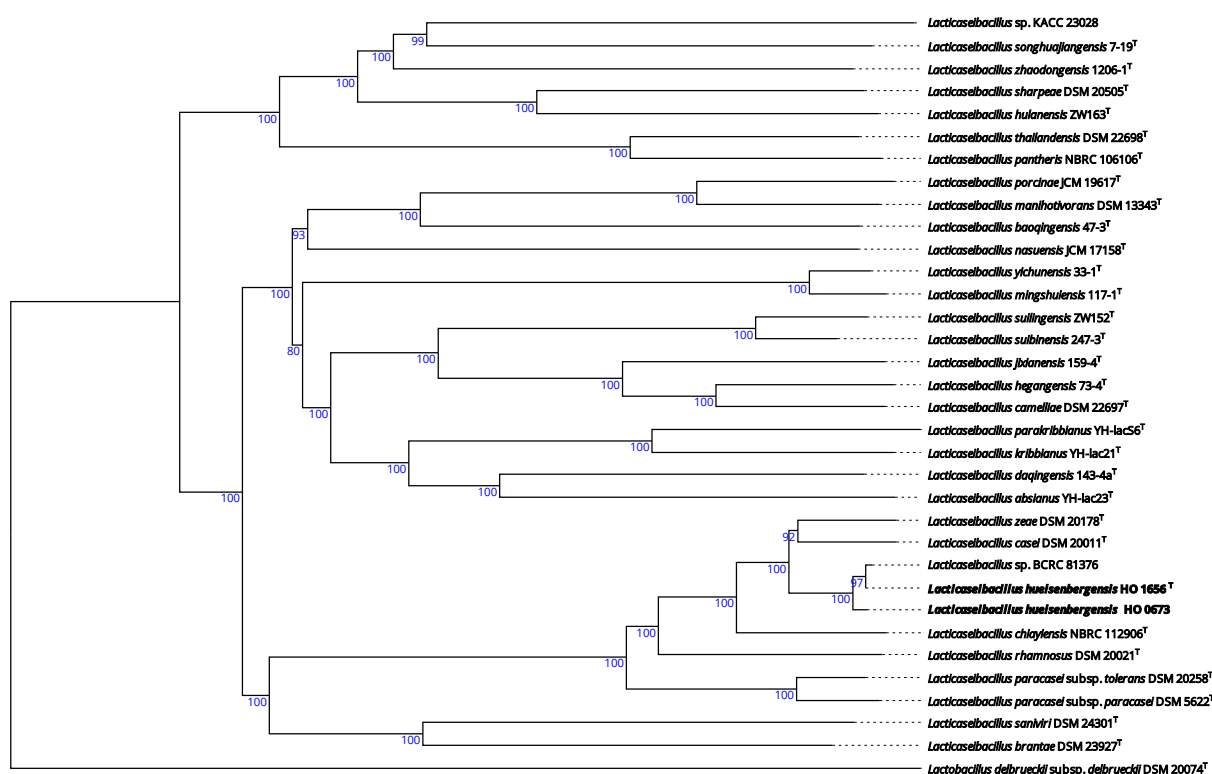


**Supplementary Figure 4.** Rep-PCR with the primers (GTG)<sub>5</sub>, ERIC1R and ERIC2 and BOXA1R.

M, 1kb DNA marker (Roth); 1, HO 0673; 2, HO 1656<sup>T</sup>; 3, *L. casei* DSM20011<sup>T</sup>; 4, *L. zeae* DSM20178<sup>T</sup>; 5, *L. chiayiensis* NBRC112906<sup>T</sup>; 6, *L. paracasei* subsp. *paracasei* DSM5622<sup>T</sup>; 7, *L. paracasei* subsp. *tolerans* DSM20258<sup>T</sup>; 8, *L. rhamnosus* DSM20021<sup>T</sup>.



**Supplementary Figure 5.** RAPD PCR of the novel isolates with the primer OPC-08, OPC-20 and OPT16. M, 1 kb DNA marker (Roth); 1, HO 0673; 2, HO 1656<sup>T</sup>.



**Supplementary Figure 6.** Phylogenetic analysis of the novel isolates' HO 1656<sup>T</sup> and HO 0673 core genome using the proteome based TYGS phylogeny. Reference strains are all valid *Lactocaseibacillus* type strains currently available at LPSN, plus all novel *Lactocaseibacillus* sp. genomes submitted to Genbank (*Lactocaseibacillus* sp. BCRC 81376 (GCF\_029589085.1) and *Lactocaseibacillus* sp. KACC 23028 (GCF\_028736235.1)), *Lactobacillus delbrueckii* subsp. *delbrueckii* DSM 20074<sup>T</sup> served as outgroup. Tree inferred with FastME 2.1.6.1 (60) from whole-proteome-based GBDP distances. The branch lengths are scaled via GBDP distance formula d5. Branch values are GBDP pseudo-bootstrap support values > 60 % from 100 replications, with an average branch support of 98.5 %. The tree was midpoint-rooted (61).