



**Figure S1.** Survival of the R-IVET library in adhesion experiment conditions. Data represent means  $\pm$  SEM from two independent experiments ( $n = 2$ ) expressed by  $\log_{10}$  CFU/mL viable bacterial population level determined by plate count.

**Table S1.** Oligonucleotides used for the constructions, verification of chromosomal R-IVET cassette, sequencing of inserts from activated clones and amplification of activated genes in various *S. thermophilus* strains

#	Sequence 5'→3'	Characteristics
1	ATCCGTAAAACCATCAATCTTAATT	Amplification of the UP#1 fragment from <i>S. thermophilus</i> LMD-9 with #2 (1204 pb) [56.9°C] Used in the first overlapping PCR with #8 (3783 pb) [†]
2	TCCCCAAGCAAAAATTGGAAC	Used with #1
3	TCCAATTTTGTCTGGGGACCAAGCAACCATT TGAG	Amplification of the promoter (prom) of kanamycin resistance gene from <i>S. thermophilus</i> TIL1193 with #4 (525 pb) [56.9°C]
4	TCTACAGTATTTAAAGATACCCC	Used with #3
5	GTATCTTTAAATACTGTAGAATAACTTCGTATA GCATACATTATACGAAGTTATAAACAGGAA*	Amplification of the promoterless kanamycin resistance gene ( <i>kanR</i> ) from <i>S. thermophilus</i> TIL1193 with #6 (1032 pb) [56.9°C]
6	GTTGCGGATGTACTTCAGAAAAG	Used with #5, #11 and #17
7	TTTCTGAAGTACATCCGCAACGTAATGTTTGG TGTAGGTAATA	Amplification of the DOWN#1 fragment from <i>S. thermophilus</i> LMD-9 with #8 (1146 pb) [62.7°C]
8	TTAAACCAATCTGTCTCGGG	Used with #1 and with #7
9	GGAACAAAAGCACCCACACT	Confirmation of <i>prom-loxP-kanR</i> presence in <i>S. thermophilus</i> STUL5002 and sequencing with #10 (4319 pb) [58°C]
10	TTAGGCTGGATGGCATAACC	Used with #9
11	CTACGAGACCTACTTAGCGC	Amplification of the UP#2 fragment from <i>S. thermophilus</i> STUL5002 with #12 (1088 pb) [62.7°C] Used in the second overlapping PCR with #6 (3421 pb) [†]
12	CTAGTTATCTACACGACGGGGAGTCAATAACT TCGTATAATGTATGC	Used with #11
13	TGACTCCCCGTCGTGTAGATAAC	Amplification of the spectinomycin resistance gene ( <i>specR</i> ) from pSET4S and sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #14 (1200 pb) [62.7°C]
14	CGCTACGATAACGCCTGTTT	Used with #13
15	AAACAGGCGTTATCGTAGCGGCTTCTGGAGCT CAATTGAA	Amplification of the <i>las</i> operon transcriptional terminator <i>Tlas</i> with #16 (252 pb) [62.7°C]
16	CTAAAGCTGACGGGGTAAAC	Used with #15
17	GTTTACCCCGTCAGCTTTAGATAACTTCGTATA GCATACATTATACGAAGTTATAAACAGGAA*	Amplification of the <i>loxP</i> -DOWN#2 fragment from <i>S. thermophilus</i> STUL5002 with #6 (947 pb) [62.7°C]
18	GCTTCTGGAGCTCAATTGAAAG	Amplification of the insert in the recombinant plasmid derived from pULNreB with #19 (variable but >460 pb) [58°C]
19	ATTCAACTTGCACCATGCCG	Used with #18
20	CGAAATAAGAGTAATATAGAG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #21 (490 pb) [58°C]
21	TAGTATCGACGGAGCCGATT	Used with #20
22	CCAGGACAATAACCTTATAGC	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #23 (536 pb) [58°C] Test for presence or absence of the <i>loxP-specR</i> fragment in the R-IVET cassette with #29 (with <i>loxP-specR</i> : 2091 pb; without <i>loxP-specR</i> : 628 pb) [58°C]
23	TCCTCCTCACTATTTTGATTAGTACC	Used with #22

24	AATAGGTACTAATCAAAATAGTGAGG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #25 (433 pb) [58°C]
25	TTTGCTTGGTAAAGCATTATGG	Used with #24
26	GGAGAGAATATTGAATGGACTAATG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #27 (455 pb) [58°C]
27	TCTAAAGCTGACGGGGTAAA	Used with #26
28	TATAGTTTACCCCGTCAGCTTTAG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #29 (445 pb) [58°C]
29	GAAAGAGCCTGATGCACTCC	Used with #22 and #28
30	CTCATGAGTGAGGCCGATG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #31 (484 pb) [58°C]
31	CAGGCTTGATCCCCAGTAAG	Used with #30
32	TGGTATGACATTGCCTTCTG	Sequencing of the <i>S. thermophilus</i> STUL5003 cassette with #33 (426 pb) [58°C]
33	TGCAAGTAAATTCACCTGTT	Used with #32
34	TTTCAAAAACAAGTGAAATCTTGA	Sequencing of the STER_0004 locus with #35 (433 pb) [58°C]
35	AAGTGACGTCCGTAATAGCTG	Used with #34
36	GCTCGAGGACTTATCCAACG	Sequencing of the STER_r0184/t0185 loci with #37 (298 pb) [58°C]
37	TACTCCGCCAGTAGGACTCG	Used with #36
38	CAAGTTTGTAGTTGGATTTTCTAGC	Sequencing of the STER_0208 locus with #39 (613 pb) [58°C]
39	GCTGTTTTGAACTTGGGCTAA	Used with #38
40	CCACAGCTAGTGTGCTTGGT	Sequencing of the STER_0303 locus with #41 (637 pb) [58°C]
41	TAGTTTCCGCCCTCAAGCGT	Used with #40
42	CGAAGGAAAAACCAAGGAGTT	Sequencing of the STER_0303 locus with #43 (713 pb) [58°C]
43	TGGATACCTTCCCAACAGAA	Used with #42
44	GAAATCCATAAATTAAGAACCTTGTC	Sequencing of the STER_0314 locus with #45 (974 pb) [58°C]
45	CGTGTCAGTCCTGCTGGTAA	Used with #44
46	AAAACCTTGTGGGGTTACTGG	Sequencing of the STER_0399 locus with #47 (873 pb) [58°C]
47	AAGAACAGCGGCAACAAAAG	Used with #46
48	CACGTTTATTCCCAACAATCA	Sequencing of the STER_0399 locus with #49 (712 pb) [58°C]
49	TGTCCGGAGAAACCAAGAAC	Used with #48
50	GCAAGAGTGATTGGCCTTGT	Sequencing of the STER_0406 locus with #51 (1103 pb) [58°C]
51	CGCTGCTTCTTAATCTGGTG	Used with #50
52	CGTTTCTTTTGGCAAGGAAG	Sequencing of the STER_0406 locus with #53 (622 pb) [58°C]
53	CGCCTTCCCAAGCTTTTTTA	Used with #52
54	CACGCGATACAGATATTAAGAAGG	Sequencing of the STER_0498/0499 loci with #55 (802 pb) [58°C]
55	CGCTACCACAAAAAGCACCT	Used with #54
56	AGATGAACTGGATGGGCTTG	Sequencing of the STER_0498/0499 loci with #57 (837 pb) [58°C]
57	GCAGGATTTTTCGCTGAGAC	Used with #56
58	TGTAGTCTTGCAAGGCTAAGGAA	Sequencing of the STER_0498/0499 loci with #59 (818 pb) [58°C]
59	CACCAAGAGTTGCCAAAAA	Used with #58
60	TGCCCTAGCTTTGATGAAT	Sequencing of the STER_0498/0499 loci with #61 (1011 pb) [58°C]
61	CCAGTGATACCAACGATTGAAA	Used with #60
62	TGTGACACCCCAATAAAAAATTA	Sequencing of the STER_0572 locus with #63 (631 pb) [58°C]
63	GGTGAAATATAAACCACGGGAAG	Used with #62
64	TCTTTCACAGGATCAGGTTGC	Sequencing of the STER_0572 locus with #65 (876 pb) [58°C]
65	TTGTGACTGAGTCGAAAATGAGA	Used with #64
66	CCCAGCTAATTCGGTTTGA	Sequencing of the STER_0582/0583 loci with #67 (929 pb) [58°C]
67	AAATCGTACAAGATACTGATTATTCG	Used with #66
68	CTACTCCCAATTAGTAGAAAGTTTTCAG	Sequencing of the STER_0582/0583 loci with #69 (623 pb) [58°C]
69	CCCTCCTTGAATGGCAGATA	Used with #68
70	ATCCCATCAAAGGCTTCGTA	Sequencing of the STER_0582/0583 loci with #71 (916 pb) [58°C]
71	TTTTGACTTCAAATATTACGTTACCT	Used with #70
72	GGCAAAGAGACCTGTTACGTG	Sequencing of the STER_0711/0712 loci with #73 (1089 pb) [58°C]
73	TTCTTCTGAGATAAAGCCATCAA	Used with #72
74	CCCTCTTTTAGAAATGTTAGACCA	Sequencing of the STER_0711/0712 loci with #75 (693 pb) [58°C]
75	GCTCTAAGATTGAGGTGAAGGTT	Used with #74
76	CGCAGCCCTCTTATCAATG	Sequencing of the STER_0758 locus with #77 (738 pb) [58°C]
77	AACAAGAGAAAAAGCAAGAGC	Used with #76
78	GGTATTGCAAATCTTCAAAGTGA	Sequencing of the STER_0807 locus with #79 (794 pb) [58°C]
79	TTTGATTCAAGTTGCTTTAAGTGG	Used with #78
80	AATCACTTTCTGGGAAGTCCA	Sequencing of the STER_0811 locus with #81 (541 pb) [58°C]
81	CATTGGCATAATCCATAGTTTCAA	Used with #80
82	CCAACAGCTTTAACCAGATTGAC	Sequencing of the STER_0901 locus with #83 (780 pb) [58°C]
83	CACCGATTCTGTAACGGAAG	Used with #82
84	AATGAATGAAGCCGCAAAAC	Sequencing of the STER_0901 locus with #85 (941 pb) [58°C]
85	TTTTTGGGGTCAGTACAAAGG	Used with #84
86	GAAAAATGTCAAGATGCCATATCT	Sequencing of the STER_0949 locus with #87 (699 pb) [58°C]
87	CCCATCTCCGATAACGACAA	Used with #86
88	AGGCTATCGGAATTGTCGAA	Sequencing of the STER_0949 locus with #89 (958 pb) [58°C]
89	GCCCTTTCGACTGTAATCATT	Used with #88
90	ACACCGCTTCCAGAAAGTGT	Sequencing of the STER_0972 locus with #91 (924 pb) [58°C]
91	GCCTACCAAGGCGTGT	Used with #90
92	CATGCTCGCAACTCATATTTAGTA	Sequencing of the STER_1045/1046/1047/1048 loci with #93 (717 pb) [58°C]

93	ACAAAAGATCCGAGCGTGC	Used with #92
94	TTTTCTCGTCAGTTGGTCTCC	Sequencing of the STER_1045/1046/1047/1048 loci with #95 (842 pb) [58°C]
95	CATCCCCACTTCCAACCTCAC	Used with #94
96	TGGAAGATGTAGGCAAGAAGC	Sequencing of the STER_1045/1046/1047/1048 loci with #97 (828 pb) [58°C]
97	CAGTAACTAAAGCTAGGACAGTTCCA	Used with #96
98	CTGTCTGGTGCCTTTTGTCC	Sequencing of the STER_1045/1046/1047/1048 loci with #99 (777 pb) [58°C]
99	CCACCGCCTTCGAGTACTAA	Used with #98
100	TCAATCAGTGAGTGCTTTTCG	Sequencing of the STER_1073/1074 loci with #101 (1015 pb) [58°C]
101	TGGGAATACGATAACTCCTCCT	Used with #100
102	GGGCAACCGTATTTTATAGGG	Sequencing of the STER_1073/1074 loci with #103 (1010 pb) [58°C]
103	GCTCTACGCGCTAAGCAGGT	Used with #102
104	GTCTGGCCGAAAAAGTGGTA	Sequencing of the STER_1196/1197 loci with #105 (879 pb) [58°C]
105	CAAGGTGATTAACCTTTTCCA	Used with #104
106	TTGGACCCCTTCACTTATGC	Sequencing of the STER_1196/1197 loci with #107 (732 pb) [58°C]
107	TTGGATTGTCCAACCTTGCTTC	Used with #106
108	TATGCGTGCATCTGGGACTA	Sequencing of the STER_1196/1197 loci with #109 (1406 pb) [58°C]
109	TGTTGTGAGTCTTACTCAAGGCTA	Used with #108
110	GGACGACTTGGTTGGCATAG	Sequencing of the STER_1319 locus with #111 (965 pb) [58°C]
111	CAAGATTTCTCAATCCTGCTACTTT	Used with #110
112	GAGGATGGAAATGTGAAAGCA	Sequencing of the STER_1356/1357 loci with #113 (696 pb) [58°C]
113	AGTTCAAGCGGAAAACTCAA	Used with #112
114	GGTATTCAATATGATGACAATTCAGA	Sequencing of the STER_1356/1357 loci with #115 (855 pb) [58°C]
115	CCTCAGAATAATTAGCTACTGAAAATC	Used with #114
116	TTTTAGGAGGGGAACCGACT	Sequencing of the STER_1356/1357 loci with #117 (1012 pb) [58°C]
117	AGGAATATCAAAAACTGAGCCACAT	Used with #116
118	TCCGATTTCATCAGCGATAG	Sequencing of the STER_1371 locus with #119 (933 pb) [58°C]
119	TCACGACGTTTGTAGTGTTC	Used with #118
120	CGCTATTGGTATGGGAGCTG	Sequencing of the STER_1371 locus with #121 (936 pb) [58°C]
121	GATTGACAACGCTCGGTCTT	Used with #120
122	GGACGTTAGCGAAGTGTCTT	Sequencing of the STER_1453 locus with #123 (822 pb) [58°C]
123	AGCCACTAGATCCCCAAGGT	Used with #122
124	TTACGGCTGGATTGAAGGTT	Sequencing of the STER_1453 locus with #125 (1044 pb) [58°C]
125	CAGCAACAAGGAAAGCCATC	Used with #124
126	TCTTGGATGAGGTGGGAAA	Sequencing of the STER_1477 locus with #127 (873 pb) [58°C]
127	TCTCCTCAATTAAGAAGTGACCT	Used with #126
128	AAAAAGCAGATTTGCGTCTAGT	Sequencing of the STER_1477 locus with #129 (1080 pb) [58°C]
129	CCTTCTCTCTGGCAAATACAAA	Used with #128
130	CCTTGGAAATTTGAGGACGTT	Sequencing of the STER_1477 locus with #131 (1076 pb) [58°C]
131	TCTCCGGTATACATATCTTTCCATT	Used with #130
132	CGCACTTCAAAATGATCGAC	Sequencing of the STER_1477 locus with #133 (1067 pb) [58°C]
133	TTTTCTTAGCACCTTTTCAATTGTC	Used with #132
134	GGGGGTATGCTGGAATTTCT	Sequencing of the STER_1477 locus with #135 (889 pb) [58°C]
135	CAATACAATGTCAGTTGTTTCCA	Used with #134
136	TCTTATCCCTTGTATCGTGTTT	Sequencing of the STER_1485 locus with #137 (569 pb) [58°C]
137	AGCGGCAACTGTTTCTATCTT	Used with #136
138	CTCGGGTGTACAATCGGTCT	Sequencing of the STER_1494 locus with #139 (1087 pb) [58°C]
139	AACCAGTTTGTCCGAAGCAC	Used with #138
140	AAAAGCTTGAAGCATGGTCAC	Sequencing of the STER_1693 locus with #141 (855 pb) [58°C]
141	TGCAAGTAAATAAGTCGATTGAA	Used with #140
142	TGCCTCGCTACTCGACTTTT	Sequencing of the STER_1693 locus with #143 (648 pb) [58°C]
143	ACCGTAGCTTATGCGTGTC	Used with #143
144	AAATTCATTCGGTGGTGAG	Sequencing of the STER_1726 locus with #145 (446 pb) [58°C]
145	CTCAGATGAGGGCATTATTTTAATC	Used with #144
146	GGTTTGTTAAGTAGTCAGTTTGATGC	Sequencing of the STER_1743 locus with #147 (593 pb) [58°C]
147	CAGCGAAGACGATATTGCAG	Used with #146
148	CATGGTGGGAAATGACTTGA	Sequencing of the STER_1814 locus with #149 (1111 pb) [58°C]
149	GCTTGCCTTTTAAATCTTCCA	Used with #148
150	GCTTTAGCACAAACGTGGAA	Sequencing of the STER_1824 locus with #151 (941 pb) [58°C]
151	ACCAATGATCCCACCAGAAA	Used with #150
152	CCAGCTAAATGGCGTACCAA	Sequencing of the STER_1859 locus with #153 (1031 pb) [58°C]
153	CAACGATAGCTGCTTGTTTCA	Used with #152
154	GCTGGGAATGTCCCATCTTA	Sequencing of the STER_1859 locus with #155 (976 pb) [58°C]
155	GCGATAATCACATCTGGTTGG	Used with #154
156	CAACACTTGGCAGGACGTA	Sequencing of the STER_1904/1905/1906 loci with #157 (945 pb) [58°C]
157	TGCAGCTTCAACTGCTTGTT	Used with #156
158	AGAAGGAAACAAATTCGCTGA	Sequencing of the STER_1904/1905/1906 loci with #159 (604 pb) [58°C]
159	TTGTTGTTACGACCAGCTTTG	Used with #158
160	CGTGTTAGACGTTTACTGGTT	Sequencing of the STER_1904/1905/1906 loci with #161 (850 pb) [58°C]
161	CGTCCAACTGGTGCTTTACC	Used with #160
162	GGTAACGCTCTTCCACTTGC	Sequencing of the STER_1904/1905/1906 loci with #163 (557 pb) [58°C]
163	CGTCCCATTTGTAGTTTCTCC	Used with #162
164	TGGAAGATAAAGAGACAAAACAAAGA	Sequencing of the STER_1951 locus with #165 (865 pb) [58°C]

165	CGCAAAGGATTTTCTTCCAA	Used with #164
166	CCAGAGGTCCAAAAACCTGT	Sequencing of the STER_1951 locus with #167 (847 pb) [58°C]
167	TGGCTTTGAAGGTATCTGCTG	Used with #166
168	CCAATATGGTACGTGGTCTGG	Sequencing of the STER_1951 locus with #169 (682 pb) [58°C]
169	GATCACTCGCAGGTTTCATCA	Used with #168
170	TTCGCTTTTGAAGTGTGACG	Sequencing of the STER_1952 locus with #171 (583 pb) [58°C]
171	TACCGTTGATACGGACGTTG	Used with #170
172	GGTGGCACCTTGATTTCTTA	Sequencing of the STER_1952 locus with #173 (973 pb) [58°C]
173	TCCACGAAACTTTTCTTGCT	Used with #172
174	TCTCTAAGTGTTCCTAGCTTCAA	Sequencing of the STER_1968 locus with #175 (1040 pb) [58°C]
175	GGCTAATTTGAGCTGGCTTTT	Used with #174
176	TTGGACGAACATAAATAGTGACG	Sequencing of the STER_B1 locus with #177 (603 pb) [58°C]
177	AGACGTCTCTTTTCGTAATTTTAAT	Used with #176

The numbers in parentheses and square brackets correspond to expected amplicons sizes and PCR annealing temperatures, respectively. †: A touchdown PCR was applied for the first 20 cycles with a 1°C decrease per cycle from 60°C to 40°C followed by a constant 42°C annealing temperature for the last 30 cycles. \*: Sequences in bold and italics correspond to loxP sites.

**Table S2.** Inserts from activated R-IVET clones identified at T0 in the different gut models

Inser t	Locus STER <sup>a</sup> (STER_RS0) b	Gene description	Location and direction compared to the gene <sup>c</sup>	Position of the insert on LMD- 9 genome (nt)	TIM mode I	Caco-2 TC7	Batch cultures of human fecal microbiot a
1	B2 (9835)	Plasmid rolling circle replication initiator protein	Promoter/sense	2082-2433		LM17/DME M	
2	0060 (0310)	Phosphoribosylaminoimidazol e carboxylase ATPase subunit	Inside/sense	48325-49378			
3	0071 (0360)	Hypothetical protein	Promoter/sense	58621-59672			
4	0095 (0480)	<i>polC</i> : DNA polymerase III catalytic subunit, PolC	Inside/sense	75226-76208		DMEM	
5	0096 (0485)	PepS aminopeptidase. Metallo peptidase. MEROPS family M29	Promoter sense	77288-77475		DMEM	
6	0113 (0565)	D-alanyl-D-alanine carboxypeptidase, penicillin- binding protein	Promoter/sense	93170-92322		LM17/DME M	
7	0120 (0595)	Hypothetical protein	Promoter/sense	98597-99644			
8	0124 (0615)	Hypothetical protein	Inside/sense	102285-102381		DMEM	
9	0125 (0620)	Hypothetical protein	Promoter/sense	102382-102740		LM17	
10	0148 (0715)	Transposase	Promoter/antisens e	122595-122799			
11	0206 (1015)	Crp-like transcriptional regulator	Promoter/sense	175028-174569		LM17/DME M	
12	0207 (1020)	Permease of the major facilitator superfamily	Promoter/sense	174822-175620		LM17	
13	0207 (1020)	Permease of the major facilitator superfamily	Inside/sense	175029-175686		DMEM	
14	0236 (1155)	Auxin efflux carrier (AEC) family permease	Promoter/sense	200160-200652		LM17	
15	0245 (1200)	CDP-diglyceride synthetase	Inside/antisense	208815-208140		LM17	
16	0384 (1865)	<i>rbfA</i> : ribosome-binding factor A	Promoter/sense	336578-337533		LM17/DME M	
17	0468 (2300)	N-formylmethionyl-tRNA deformylase	Promoter/sense	414741-415335		LM17/DME M	
18	0509 (1030)	Glycopeptide antibiotics resistance protein	Promoter/sense	457780-457906		LM17	
19	0531 (2605)	Ammonia permease	Promoter/sense	477410-477541		LM17	
20	0537 (2635)	N-acetylglucosamine 6- phosphate deacetylase	Promoter/sense	481688-482351		LM17	
21	0573 (2825)	Predicted protein	Promoter/sense	517470-518024			
22	0583 (2870)	Response regulator of the LytR/AlgR family	Promoter/sense	531843-530990			
23	0611 (3000)	Glycosyl transferase, family 1	Inside/sense	555151-556148			
24	0612 (3005)	Hypothetical protein	Promoter/sense	556224-556595		DMEM	

25	0623 (3055)	Dihydrofolate reductase	Promoter/sense	566220-566539			
26	0623 (3055)	Dihydrofolate reductase	Promoter/sense	566220-566311		LM17	
27	0660 (3255)	Methenyltetrahydrofolate cyclohydrolase	Promoter/sense	598595-598901		LM17	
28	0711 (3500)	CRISPR-associated protein, Cas2 family	Promoter/sense	647294-647916			
29	0744 (3650)	Uncharacterized conserved protein	Promoter/sense	675683-675866			
30	0747 (3665)	Type I site-specific restriction-modification system, R (restriction) subunit or related helicase	Inside/sense	679498-680421			
31	0761 (3735)	<i>hola</i> : DNA polymerase III subunit delta	Promoter/sense	694068-694911			
32	0813 (4000)	Uncharacterized conserved phage related protein	Promoter/sense	747866-747426			
33	0830 (4075)	Hypothetical protein	Promoter/sense	755652-756572			
34	0872 (4310)	Enzyme related to GTP cyclohydrolase I	Promoter/sense	802930-803563		LM17	
35	0885 (4380)	Cystathionine beta-synthase (acetylserine-dependent)	Promoter/sense	817241-817531			
36	0948 (4690)	Ferrochelatase	Promoter/sense	877316-878238		LM17/DME M	
37	0949 (4595)	Threonine dehydrogenase or related Zn-dependent dehydrogenase	Promoter/sense	879316-879396		LM17	
38	0950/0951 (-/4700)	Hypothetical protein/pseudogene	Promoter/sense	879949-880988		LM17	
39	0981 (4840)	<i>pyrE</i> : Orotate phosphoribosyltransferase	Promoter/sense	905098-905735		DMEM	
40	1012 (4990)	Lysyl aminopeptidase. Metallo peptidase. MEROPS family M01	Inside/sense	934009-935056			
41	1013 (4995)	<i>xerS</i> : Site-specific tyrosine recombinase XerS	Promoter/sense	936928-935885		LM17/DME M	
42	1043 (5175)	Pseudogene	Promoter/sense	967939-968905		LM17	
43	1102 (5460)	<i>prfB</i> : peptide chain release factor 2	Promoter/sense	1019356-1018918			
44	1166 (5755)	Putative translation factor (SUA5)	Promoter/sense	1078437-1078262		LM17/DME M	
45	1197 (5905)	Cell division membrane protein	Promoter/sense	1106940-1106003			
46	1238 (6110)	Uridine kinase	Promoter/sense	1144304-1143294		LM17	
47	1280 (6320)	<i>urvC</i> : excinuclease ABC subunit C	Inside/sense	1190055-1189837		LM17/DME M	
48	1293 (6375)	ABC-type multidrug transport system, ATPase component	Promoter/antisense	1201333-1202291			
49	1298 (-)	Hypothetical protein	Promoter/sense	1206867-1207715			
50	1320 (6520)	Predicted ring-cleavage extradiol dioxygenase	Inside/antisense	1228239-1228286		DMEM	
51	1356 (6690)	Radical SAM superfamily enzyme	Inside/sense	1260895-1260614		LM17/DME M	
52	1400 (6885)	Predicted hydrolase of the HAD superfamily	Promoter/sense	1310880-1309989			
53	1406 (6915)	ABC-type dipeptide/oligopeptide/nickel transport system, ATPase component	Inside/sense	1314940-1314560		LM17/DME M	
54	1408 (6925)	ABC-type dipeptide/oligopeptide/nickel transport system, permease component	Inside/sense	1317075/1316209			
55	1453 (7150)	Aminotransferase	Promoter/sense	1361583-1361200		DMEM	

56	1469 (7225)	Uncharacterized protein, possibly involved in aromatic compounds catabolism	Promoter/sense	1374989-1374939	LM17/DME M	
57	1518 (7470)	SSU ribosomal protein S5P alanine acetyltransferase	Promoter/sense	1422905-1422091	LM17	
58	1522 (7490)	1-acyl-sn-glycerol-3-phosphate acyltransferase	Promoter/sense	1428562-1427572		
59	1539 (7565)	ABC-type amino acid transport/signal transduction system, periplasmic component/domain	Promoter/sense	1443950-1443474	DMEM	
60	1579 (7760)	Hypothetical protein	Inside/antisense	1479868-1480392	LM17/DME M	
61	1585 (7790)	Predicted GTPase	Inside/antisense	1484613-1485240		
62	1586 (7795)	Predicted hydrolase of the HAD superfamily	Promoter/sense	1486730-1486347	LM17	
63	1591 (7815)	<i>gatA</i> : aspartyl/glutamyl-tRNA(Asn/Gln) amidotransferase subunit A	Inside/sense	1490454-1490313	DMEM	
64	1597 (7840)	ABC-type metal ion transport system, periplasmic component/surface antigen	Promoter/sense	1495479-1495063		
65	1643 (8060)	Hypothetical protein	Promoter/sense	1535445-1535290	LM17	
66	1664 (8140)	Superfamily II DNA and RNA helicase	Promoter/sense	1554241-1553929	LM17	
67	1684 (8235)	Response regulator of the LytR/AlgR family	Promoter/sense	1576456-1576201	LM17/DME M	
68	1736 (8480)	DNA polymerase I	Promoter/sense	1625236-1624238	LM17	
69	1774 (8680)	Mg-dependent DNase	Promoter/sense	1659581-1659384	LM17/DME M	
70	1846 (9020)	Membrane carboxypeptidase (penicillin-binding protein)	Promoter/sense	1723432-1723281	DMEM	
71	1862 (9110)	Pseudogene	Promoter/sense	1743723-1742676	LM17	
72	1866 (9140)	Hypothetical protein	Promoter/sense	1745938-1745578		
73	1895 (9285)	LSU ribosomal protein L5P	Promoter/sense	1763621-1763389		
74	1912/1911 (9365/9370)	Protein-tyrosine-phosphatase/Uncharacterized protein conserved in bacteria	Promoter/sense	1773889-1772913		
75	P1: 0061 (0315)	Pseudogene	Inside/antisense	50147-50034	LM17/DME M	
	P2: -	No gene identified	-	428255-428540		
76	P1: 0107 (0535)	Predicted membrane protein	Inside/antisense	85074-85135	LM17/DME M	
	P2: 1980 (9680)	Hypothetical protein	Promoter/sense	1834023-1833850		
	P3: 0338 (-)	Hypothetical protein	Promoter/sense	292729-292793		
77	P1: 0157 (0760)	Uncharacterized conserved protein	Inside/sense	129818-129958	LM17	
	P2: 0633 (3105)	DNA topoisomerase IV subunit B	Inside/sense	575858-576757		
78	P1: 0220 (1070)	Cysteine desulfurase	Inside/sense	186671-187317	LM17/DME M	
	P2: 1772 (8670)	<i>ksgA</i> : dimethyladenosine transferase	Promoter/sense	1658295-1657961		
79	P1: 0267 (1295)	Pseudogene	Inside/antisense	227033-227182	LM17/DME M	
	P2: B2 (9835)	Plasmid rolling circle replication initiator protein	Inside/antisense	2435-2300		
80	P1: 0307 (1490)	<i>scpA</i> : segregation and condensation protein A	Inside/sense	264475-264593	DMEM	

81	P2: 1111 (5495)	Predicted hydrolase of the HAD superfamily	Promoter/sense	1025666-1026525	LM17	
	P1: 0311 (1510)	Trk-type K <sup>+</sup> transport system, membrane component	Promoter/antisense	268092-268381		
	P2: 0198 (0970)	5'-nucleotidase/2',3'-cyclic phosphodiesterase or related esterase	Inside/antisense	164148-164035		
82	P3: 0662 (3265)	Cell division protein FtsI/penicillin-binding protein 2	Promoter/sense	600539-601100	LM17	
	P1: 0351 (1710)	Trk-type K <sup>+</sup> transport system, membrane component	Inside/antisense	307122-307554		
	P2: 0839 (4130)	Pseudogene	Inside/antisense	768120-767545		
83	P1: 0388 (1890)	Membrane protein involved in polysaccharide transport	Inside/sense	342566-342669	LM17/DME M	
	P2: 0744 (3650)	Uncharacterized conserved protein	Promoter/sense	675681-675865		
84	P1: 0528 (2590)	Regulator of signaling phosphorelay	Promoter/antisense	475645-474908	LM17	
	P2: -	No gene identified	-	832730-832547		
85	P1: 0694 (3415)	Malate dehydrogenase (NAD)	Inside/antisense	630957-630878	DMEM	
	P2: 1123 (5555)	UDP-N-acetylglucosamine 1-carboxyvinyltransferase	Promoter/sense	1037911-1036955		
86	P1: 0751 (3685)	Hypothetical protein	Inside/antisense	684811-184699	LM17/DME M	
	P2: 1513 (7445)	NAD-dependent DNA ligase	Inside/sense	1418027-1417943		
	P3: 1524 (7500)	Hypothetical protein	Promoter/sense	1428673-1428787		
87	P1: 0761 (3735)	<i>hola</i> : DNA polymerase III, delta subunit	Promoter/sense	694511-694643	LM17	
	P2: 0699 (3440)	Tellurite resistance protein or related permease	Inside/antisense	636263-636379		
88	P1: 0777 (3810)	Cell division protein FtsZ	Inside/antisense	710969-710860	DMEM	
	P2: 1454 (7155)	ABC-type amino acid transport/signal transduction system, periplasmic component/domain	Promoter/antisense	1362197-1363049		
89	P1: 1103 (-)	Hypothetical protein	Promoter/sense	1019044-1018908	LM17	
	P2: 0375 (1820)	Xanthine/uracil/vitamin C permease	Inside/sense	329323-329405		
	P3: 1861 (9105)	Pseudogene	Inside/sense	1742078-1741907		
90	P1: 1134 (5600)	Hypothetical protein	Inside/antisense	1047111-1047304		
	P2: 0289 (1400)	<i>greA</i> : transcription elongation factor GreA	Promoter/sense	251808-252550		
91	P1: 1139 (5625)	GAF domain-containing protein	Promoter/sense	1054731-1054392	LM17/DME M	
	P2: 1715 (8380)	Translation elongation factor P (EF-P)	Inside/sense	1606151-1606065		
92	P1: 1379 (6790)	Glutamate-cysteine ligase	Inside/antisense	1287692-1287449	LM17/DME M	
	P2: 0336 (1635)	NUDIX family hydrolase	Promoter/antisense	289592-288847		
93	P1: 1400 (6885)	Predicted hydrolase of the HAD superfamily	Inside/sense	1310087/1309642	LM17/DME M	
	P2: 0574 (2830)	Uncharacterized conserved phage related protein	Inside/sense	518690-519174		
94	P1: 1413 (6950)	Transposase	Inside/antisense	1324280-1324120		
	P2: 0252 (1225)	<i>groES</i> : co-chaperonin GroES (HSP10)	Promoter/sense	215497-215643		

95	P3: 0252 (1225)	<i>groES</i> : co-chaperonin GroES (HSP10)	Promoter/sense	215675-215798		
	P1: 1461 (7190)	ABC-type amino acid transport system, periplasmic and permease components	Inside/antisense	1369441-1369027		
	P2: 1710 (8355)	Sucrose PTS, EIIBCA	Inside/sense	1600881-1600629		
96	P3: 1489 (7320)	Phosphoserine aminotransferase	Promoter/sense	1395106-1394818		
	P1: 1534 (7550)	UDP-N-acetylmuramoyl-tripeptide--D-alanyl-D-alanine ligase	Promoter/sense	1441347-1440845		
	P2: 0862 (4250)	Uncharacterized protein conserved in bacteria	Inside/sense	796404-796597		
97	P3: 0864 (4260)	<i>asnC</i> : asparaginyl-tRNA synthetase	Inside/sense	798722-799012		
	P1: 1676 (8195)	Glutamate 5-kinase	Promoter/sense	1564089-1563756	LM17/DME M	
	P2: 1797 (8780)	<i>rplA</i> : LSU ribosomal protein L1P	Inside/antisense	1681062-16811278		
98	P1: 1682 (8225)	DNA helicase/exodeoxyribonuclease V, subunit B	Promoter/sense	1574798-1574554	LM17/DME M	
	P2: 0389 (1895)	Pseudogene	Promoter/antisense	343134-343016		
	P1: 1684 (8235)	Response regulator of the LytR/AlgR family	Promoter/sense	1576456-1576301		
100	P2: 0383 (1860)	<i>infB</i> : bacterial translation initiation factor 2 (bIF-2)	Inside/antisense	336251-335993		
	P1: 1689 (8255)	Uncharacterized protein conserved in bacteria (Cof family protein)	Inside/sense	1578827-1578740	LM17/DME M	
	P2: 1000 (4930)	<i>truB</i> : tRNA pseudouridine synthase B	Promoter/sense	922786-922977		
101	P1: 1690 (8260)	Asparaginase	Inside/antisense	1581011-1580885	LM17	
	P2: 0165 (0800)	<i>truA</i> : Pseudouridine synthase A	Inside/antisense	139087-138850		
	P3: -	No gene identified	-	1270201-1270550		
102	P1: 1709 (8350)	Fructokinase	Inside/sense	1599180-1599064	DMEM	
	P2: 1184 (5845)	<i>xseA</i> : exodeoxyribonuclease VII large subunit	Promoter/sense	1094776-1093928		
	P1: 1746 (8530)	Acetyltransferase	Inside/antisense	1635078-1635212		
104	P2: 1180 (5825)	Transcriptional regulator, ArgR family	Promoter/sense	1091139-1090473		
	P1: 1827 (8925)	Glutamyl aminopeptidase. Metallo peptidase. MEROPS family M42	Promoter/sense	1702403-1702595	DMEM	
	P2: 0760 (3730)	Phosphoenolpyruvate carboxylase	Inside/antisense	692925-692835		
105	P1: 1857 (9070)	L-threonine synthase	Inside/sense	1736178-1735869		
	P2: 0761 (3735)	<i>holA</i> : DNA polymerase III, delta subunit	Promoter/sense	694250-694640		
	P1: 1889 (8255)	<i>rpmD</i> : 50S ribosomal protein L30	Inside/antisense	1760360-1760519	LM17/DME M	
107	P2: 1555 (7645)	Chorismate mutase	Promoter/sense	1458978-1458361		
	P1: 1946 (9515)	Hypothetical protein	Promoter/sense	1804618-1804321	LM17/DME M	
	P2: 0126 (0625)	Ketopantoate reductase	Inside/antisense	103934-103277		
108	P1: 1969 (9630)	Predicted membrane protein	Inside/antisense	1820741-1820903		



109	P2: 1992 (9740)	Inosine-5'-monophosphate dehydrogenase	Promoter/antisens e	1845233- 1846062	DMEM	
	P1: 1969 (9630)	Predicted membrane protein	Inside/antisense	1820633- 1820859		
	P2: 0595 (2930)	Pseudogene	Promoter/antisens e	541106-540512		
110	P1: -	No gene identified	-	429681-429550	DMEM	
	P2: 0787 (3860)	<i>rpmE2</i> : LSU ribosomal protein L31P	Promoter/sense	720571-720098		

(<sup>a</sup>): locus annotation dating from 01-28-2014; (<sup>b</sup>): locus annotation dating from 07-30-2015 on NCBI; (<sup>c</sup>): See Fig. 5 for term explanations. P1, P2 and P3: when a single insert has overall or part of its sequence which aligns with different (two or three) regions of LMD-9 genome.

**Table S3.** Allele variability of R-IVET activated genes identified in TIM-1 and fecal batch cultures

		Gene											Protein												
	<i>S. thermophilus</i> classes <sup>c</sup>	6	6	6	5	4	4	3	2	2	2	1	6	6	6	5	4	4	3	2	2	2	1		
	Resistance level to GI stresses <sup>d</sup>	High level	High level	High level	High level	Intermediate level	Intermediate level	Intermediate level	Low level	Low level	Low level	High level	High level	High level	High level	High level	Intermediate level	Intermediate level	Intermediate level	Low level	Low level	Low level	High level	Signature High Level <sup>e</sup>	Signature Low Level <sup>f</sup>
Locus STER_ <sup>a</sup> (STER RS) <sup>b</sup>	Gene description	LMD-9 <sup>g</sup>	PB180	PB302	EBLST20	LMG18311	CNRZ160	PB385	EBL308	ST14	CNRZ21	EBL385	LMD-9 <sup>g</sup>	PB180	PB302	EBLST20	LMG18311	CNRZ160	PB385	EBL308	ST14	CNRZ21	EBL385		
0208 (01025)	30S ribosomal protein S15	1	1	1	1	2	2	1	3	4	4	1	a	a	a	a	a	a	a	a	a	a	a	a	a
0303 (01470)	Non-canonical purine NTP pyrophosphatase	1	2	2	2	3	4	2	5	6	1	2	a	b	b	b	c	c	b	d	e	a	b	b	a/d/e/
0399 (01945)	Branched-chain amino acid ABC-type transport system, permease component	1	2	2	2	3	4	2	5	6	3	7	a	a	a	a	a	a	a	a	a	a	b	a	a
0498 (02445)	Predicted amydohydrolase	1	2	2	2	3	N D	2	N D	4	3	5	a	b	b	b	c	ND	b	ND	d	c	b	b	c/d
0499 (02450)	Transaminase	1	1	1	1	2	3	1	4	5	2	6	a	a	a	a	b	a	a	c	d	b	e	a	b/c/d
0572 (02820)	ABC permease transporter	1	1	1	1	-	-	1	-	-	-	2	a	a	a	a	-	-	a	-	-	-	b	a	-
0901 (04465)	LysR family transcriptional regulator	1	1	1	1	2	3	1	4	3	2	5	a	a	a	a	b	a	a	b	a	b	c	a	b
0949 (04695)	Zn-dependant alcohol dehydrogenase	1	1	1	1	2	3	1	4	3	2	3	a	a	a	a	b	a	a	c	a	b	a	a	a/b/c
0972 (04800)	CRISPR-associated endoribonuclease Cas6	1	1	1	1	2	-	1	3	4	2	5	a	a	a	a	b	-	a	c	c	b	d	a	c
1046 (5190)	Hypothetical protein	1	1	1	1	2	3	1	4	5	2	4	a	a	a	a	b*	c**	a	d**	b*	b*	d**	a	b*
1047 (05195)	Predicted unusual protein kinase	1	2	2	2	3	4	2	5	6	3	ND	a	b	b	b	c	d	b	e	f	c	ND	b	c/e/f
1048 (05200)	Hypothetical protein (phasin protein superfamily)	1	1	1	1	2	3	1	4	5	2	ND	a	a	a	a	b	b	a	c	d	b	ND	a	b/c/d
1319 (06515)	Hypothetical protein (nitroreductase-like protein family)	1	2	2	2	3	4	5	6	7	8	9	a	b	b	b	c	d	b	e	f	g	e	b	e/f/g
1356 (06690)	KxxxW cyclic peptide radical SAM maturase	1	1	1	1	-	-	1	-	-	-	1	a	a	a	a	-	-	a	-	-	-	a	a	-
1357 (10575)	Hypothetical protein	1	1	1	1	-	-	1	-	-	-	1	a	a	a	a	-	-	a	-	-	-	a	a	-
1693 (08275)	Transcriptional regulator (helix-turn-helix XRE-family like protein)	1	1	1	1	-	-	1	-	-	-	-	a	a	a	a	-	-	a	-	-	-	-	a	-

<b>1814 (08865)</b>	2,3,4,5-tetrahydropyridine-2,6-dicarboxylate N-acetyltransferase	1	1	1	1	2	1	1	3	1	2	4	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a
<b>1824 (08910)</b>	EMAP domain-containing protein	1	1	1	1	2	3	1	4	5	6	7	a	a	a	a	a	a	a	a	b	a	a	a	a	a	a	a	a
<b>1726 (08440)</b>	30S ribosomal protein S18	1	1	1	1	1	1	1	1	1	1	1	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a
<b>1905 (09335)</b>	LSU ribosomal protein L23P	1	1	1	1	1	1	1	1	2	1	1	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a	a
<b>1906 (09340)</b>	50S ribosomal protein L4	1	1	1	1	2	2	1	1	3	2	1	a	a	a	a	a	a	a	a	b	a	a	a	a	a	a	a	a
<b>1951 (09535)<sup>h</sup></b>	Histidyl-tRNA synthetase	1	1	1	1	2	3	1	4	2	5	6	a	a	a	a	b	c	a	d	b	b	a	a	a	a	a	a	a
<b>r0184 (00895)<sup>i</sup></b>	rRNA-5S ribosomal RNA	1	1	1	1	1	1	1	1	1	1	1																	
<b>t0185 (00900)<sup>i</sup></b>	tRNA-Asn	1	1	1	1	1	1	1	1	1	1	1																	

(<sup>a</sup>): locus annotation from 01-28-2014; (<sup>b</sup>): locus annotation from 07-30-2015 on NCBI; (<sup>c</sup>): according to Junjua *et al.*, 2016; (<sup>d</sup>): GIT stresses include acid pH, bile salts and oxidative stresses; (<sup>e</sup>): type of proteins found in strains exhibiting a high level of resistance against acid stress; (<sup>f</sup>): type of proteins found in strains exhibiting a low level of resistance against acid stress (<sup>g</sup>): LMD-9 used as a reference strain for comparison; (<sup>h</sup>): gene identified in a previous study (Uriot *et al.*, 2016); (<sup>i</sup>): no protein for these genes. (\*): stop codon after 20 amino acids; (\*\*): no stop codon identified. (-): no PCR amplicon; ND: PCR amplicons but with no analysable sequence.

**Table S4.** Allele variability of R-IVET activated genes identified in Caco-2 TC7 cells

		Gene								Protein							
		<i>S. thermophilus</i> classes <sup>c</sup>															
		6	6	5	5	4	4	2	1	6	6	5	5	4	4	2	1
		High adhesion level	Intermediate adhesion level	High adhesion level	High adhesion level	Low adhesion level	High adhesion level	Low adhesion level	Intermediate adhesion level	High adhesion level	Intermediate adhesion level	High adhesion level	High adhesion level	Low adhesion level	High adhesion level	Low adhesion level	Intermediate adhesion level
	Adhesion capacity to HT29-MTX intestinal cells																
Locus	Gene description	LMD-9 <sup>d</sup>	PB18O	ST88	EBLST20	PB5MJ	LMG18311	CNRZ21	EBL385	LMD-9 <sup>d</sup>	PB18O	ST88	EBLST20	PB5MJ	LMG18311	CNRZ21	EBL385
STER_ <sup>a</sup>																	
(STER_RS) <sup>b</sup>																	
0004 (00020)	Hypothetical protein	1	1	1	1	1	1	2	-	a	a	a	a	a	a	b	-
0314 (01525)	Membrane protein	1	1	1	1	1	2	3	4	a	a	a	a	a	b	b	b
0406 (01985)	Competence protein	1	1	1	1	1	2	2	3	a	a	a	a	a	b	b	c
0582 (02865)	Predicted signal transduction protein with C-terminal HATPase domain	1	1	1	1	1	2	1	1	a	a	a	a	a	b	a	a
0583 (02870)	DNA-binding response regulator	1	1	1	1	1	2	2	1	a	a	a	a	a	b	b	a
0711 (03500)	CRISPR-associated protein, Cas2 family	1	2	1	2	1	3	3	ND	a	b	a	b	a	c	c	ND
0712 (03505)	Uncharacterized conserved protein	1	ND	1	2	1	3	4	ND	a	ND	a	b	a	c	d	ND
0758 (03720)	Membrane protein	1	1	1	1	1	2	3	4	a	a	a	a	a	b	c	d
0807 (03965)	Hypothetical protein	1	2	1	2	1	3	4	-	a	b*	a	b*	a	c**	d*	-
0811 (03990)	Transposase	1	1	1	1	1	-	-	1	a	a	a	a	a	-	-	a
1196 (05900)	IS5 family transposase	1	1	1	1	1	-	-	-	a	a	a	a	a	-	-	-
1197 (05905)	Rod shape-determining protein RodA	1	1	1	1	1	2	3	4	a	a	a	a	a	b	c	a
1371 (06720)	Galactokinase	1	ND	1	2	1	3	4	5	a	ND	a	b	a	b	b	c
1453 (07150)	Glutamine amidotransferase	1	1	1	1	1	2	2	3	a	a	a	a	a	b	b	c
1477 (07260)	Type II CRISPR RNA-guided endonuclease Cas9	1	1	1	1	1	-	-	2	a	a	a	a	a	-	-	b
1485 (07300)	Arsenate reductase family protein	1	1	1	1	1	1	2	2	a	a	a	a	a	a	b	b
1494 (07350)	Spermidine/putrescine ABC transporter permease	1	1	1	1	1	2	3	1	a	a	a	a	a	b	c	a
1743 (08515)	Branched-chain amino acid transporter	1	1	1	1	1	2	2	3	a	a	a	a	a	b	b	b
1968 (09625)	Hypothetical protein	1	1	1	1	1	2	2	1	a	a	a	a	a	b	b	a
B1 (09830)	Acid-shock protein	1	-	1	1	1	-	-	-	a	-	a	a	a	-	-	-

(<sup>a</sup>): locus annotation dating from 01-28-2014; (<sup>b</sup>): locus annotation dating from 07-30-2015 on NCBI; (<sup>c</sup>): according to Junjua *et al.*, 2016; (<sup>d</sup>): LMD-9 used as a reference strain for comparison. (\*): an extra tryptophan at the end of sequence; (\*\*): stop codon after 60 amino acids. (-): no PCR amplicon; ND: No analysable sequence.