

JOURNAL OF FOOD SCIENCE & TECHNOLOGY

Supplementary Material For:

**Extracellular production of *Streptomyces ladakanum*
transglutaminase in a food-grade strain, *Bacillus subtilis***

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Contents:

Table S1: Primers used in this study.

Table S2: Sequences of signal peptides.

Fig. S1: Construction of plasmids carrying different signal peptides.

Fig. S2: PCR amplification of the encoding gene of *S. ladakanum* TG (A) and

SDS-PAGE analysis of *S. ladakanum* TG (B).

Table S1 Primers used in this study.

Name	Oligonucleotides (5' to 3')
TG-F*	CGGT <u>ACCCGGGGCC</u> ACCGGCAGTGGCAGTGG
TG-R*	<u>CTCTAGAGG</u> ATCGAGCGGCCAGCCCTGTGTCACCT
ProTG-F*	CCTCGAGCTCGGT <u>ACCCGGGGCC</u> ACCGGCAGTGGCAGTGGCAGCG
ProTG-R	CAGCCCTCCTGGTACCGCTATCACTTTAGGGGGCCCGGAAGGACGGACCG
CD-F	AGGAGGCGCAACTCAAGCTTTTGCCGATCCTCTAGAGGACTCCGACGAGC
CD-R*	AAGCTAGCTTGCATGCCTGCAGGTCGAGGCCAGCCCTGTGTCACCTTGTC
SacB-F	CGGTCCGTCTTCCGGGGCCCCCTAAAGTGATAGCGGTACCAGGAGGGCTG
SacB-R	GCTCGTCGGAGTCCTCTAGAGGATCGGCAAAAGCTTGAGTTGCGCCTCCT
DSD-F#	GTCTTCCGGGGCCCC <u>GCAGCAGC</u> AGAGCGGGTGACTCCTCCCGC
DSD-R#	GCGGGAGGAGTCACCCGCTCT <u>TGCTGCTG</u> CGGGGGCCCCGGAAGGAC
ΔPS-F	CTTCGAGCGCCGGTTTCCGGGGCCCCGA
ΔPS-R	TCGGGGGGCCCCGGAACCGGCGCTCGAAG
PS-F#	AGCGCCGGT <u>GCAGC</u> ATTCCGGGGCCCC
PS-R#	GGGCCCCGGAAT <u>TGCTGC</u> ACCGGCGCTC
SGS-F#	GGCAGTGGC <u>GCAGCAGC</u> AGGCACCGGGG
SGS-R#	CCCCGGTGCT <u>TGCTGCTG</u> CGCCACTGCCGGT
PD-F#	GCCACCGGCAGTGGC <u>GCAGCAGC</u> AGGCACCGGGGAAGAG
PD-R#	CTCTTCCCCGGTGCCT <u>TGCTGCTG</u> CGCCACTGCCGGTGGC
WapA-F*	TGAAC <u>GATGAACATCGGATCC</u> ATGAAAAAAGAAAGAGGCGAA
WapA-R*	TGGCCCCGGGTACCGAGCTCGAGGCTAGTACATCGGCTGGCA
EpR-F*	CATCT <u>GGATCC</u> ATGAAAAACATGTCTTGCAAAC
EpR-R*	TGGCCCCGGGTACCGAGCTCGACGCATGAGCGAGAGGGCCTA
NucB-F*	TCT <u>GGATCC</u> ATGAAAAAATGGATGGCAGGCCT
NucB-R*	GGCCCCGGGTACCGAGCTCGACGAAGATGCGCCTTGGATCTG
YncM-F*	TCT <u>GGATCC</u> ATGGCGAAACCACTATCAAAAGGGG
YncM-R*	GGCCCCGGGTACCGAGCTCGAGGCGTCTGCCGCGGGTAAA
YhcR-F*	TCT <u>GGATCC</u> ATGCTGTCTGTGCGAAATGATAA
YhcR-R*	GGCCCCGGGTACCGAGCTCGAAGCTTCGAACGTGTACATTAC
WprA-F*	TCT <u>GGATCC</u> ATGAAACGCAGAAAATTCAGCT
WprA-R*	GGCCCCGGGTACCGAGCTCGACGCTGCAGCTTTGGTTCCCCGG
AmyE-F*	TCT <u>GGATCC</u> AGATGTTTGCAAACGATTCAAAC
AmyE-R*	TGGCCCCGGGTACCGAGCTCGAGGCACTCGCAGCCGCCGG
LytD-F*	TCT <u>GGATCC</u> ATGAAAAAGAGACTAATCGCACCT
LytD-R*	GGCCCCGGGTACCGAGCTCGAGGCCTGGGCAGAACCAGACAT
PenP-F*	TCT <u>GGATCC</u> ATGAAGTTGAAAATAAAGCGTCAA
PenP-R*	TGGCCCCGGGTACCGAGCTCGAGGCTTCGGCATGTGTTGAGT

AbnA-F* TCTGGATCCCAATGAAAAAGAAAAAACATGG
 AbnA-R* GGCCCCGGGTGC GGGAGCAGCAGAAGTGAAT
 NprB-F* ACATGGATCCATGCGCAACTTGACCAAGAC
 NprB-R* GGCCCCGGGAGCTGAGGCATGTGTTACAAAAAC
 BglS-F* TCTGGATCCCAATGCCTTATCTGAAACGAGTG
 BglS-R* TGGCCCCGGGAGCTGAGGCAGTAGCAGTGACTG
 MotB-F* TCTGGATCCCAATGGCGAGAAAAAGAAGAAGA
 MotB-R* GTGGCCCCGGGGCTGCTCGCGTACAGCACAATAAAC
 LipB-F* TCTGGATCCCAATGAAAAAGTACTTATGGCATT
 LipB-R* GGTGGCCCCGGGAGCTTTTGCGCCAGACGGCG
 LipA-F* ATCTGGATCCCAATGAAATTTGTA AAAAGAAGG
 LipA-R* GGCCCCGGGGGCTTTTGCTGACGGCTGCAACGC

* Restriction sites and #mutagenesis sites are underlined.

Table S2 Sequences of signal peptides.

Name	Oligonucleotides (5' to 3')
SacB	ATGAACATCAAAAAGTTTGCAAACAAGCAACAGTATTAACCTTTACTACC GCACTGCTGGCAGGAGGCGCAACTCAAGCTTTTGC
WapA	ATGAAAAAAGAAAGAGGCGAAACTTTAAAAGGTTTCATTGCAGCATTTTTA GTGTTGGCTTTAATGATTTTCATTAGTGCCAGCCGATGTACTAGCC
EpR	ATGAAAAACATGTCTTGCAAACCTTGTTGTATCAGTCACTCTGTTTTTCAGTT TTCTCACCATAGGCCCTCTCGCTCATGCG
NucB	ATGAAAAAATGGATGGCAGGCCTGTTTCTTGCTGCAGCAGTTCTTCTTTGTT TAATGGTTCGCCAACAGATCCAAGGCGCATCTTCG
YncM	ATGGCGAAACCACTATCAAAGGGGGAATTTGGTGAAAAAAGTATTGATT GCAGGTGCAGTAGGAACAGCAGTTCTTTTCGGAACCCTTTCATCAGGTATA CCAGGTTTACCCGCGGCAGACGCC
YhcR	ATGCTGTCTGTGCAAATGATAAGCAGACAAAATCGTTGTCATTATGTGTATA AGGGAGGAAATATGATGAGGCGTATTCTGCATATTGTGTTGATCACGGCATT AATGTTCTTAAATGTAATGTACACGTTTCGAAGCT
WprA	ATGAAACGCAGAAAATTCAGCTCGGTTGTGGCGGCAGTGCTTATTTTTGCA CTGATTTTCAGCCTTTTTTCTCCGGGAACCAAAGCTGCAGCG
AmyE	ATGTTTGCAAACGATTCAAACCTCTTTACTGCCGTTATTCGCTGGATTTT TATTGCTGTTTCATTTGGTTCTGGCAGGACCGGCGGCTGCGAGTGCC
LytD	ATGAAAAAGAGACTAATCGCACCTATGCTTCTATCCGCCGCGTCCCTTGCCCT TTTTTGCCATGTCTGGTTCTGCCAGGCC
PenP	ATGAAGTTGAAAATAAAGCGTCAATAAAATTCGGAATATGTGTTGGGCTT TTATGTTTAAGCATTACTGGTTTCACACCTTTTTTCAACTCAACACATGCCG AAGCC
AbnA	ATGAAAAAGAAAAAACATGGAAACGCTTCTTACACTTTTCGAGTGCAGC TCTGGCTGCAGGTTTGATATCACTTCTGCTGCTCCCGCA
NprB	ATGCGCAACTTGACCAAGACATCTCTATTACTGGCCGGCTTATGCACAGCG GCCCAAATGGTTTTTGTAACACATGCCTCAGCT
BglS	ATGCCTTATCTGAAACGAGTGTTGCTGCTTCTTGTCAGTGGATTGTTTATGA GTTTGTGTTGCAGTCACTGCTACTGCCTCAGCT
MotB	ATGGCGAGAAAAAGAAGAAGAAGCATGAGGACGAGCACGTTGATGAAT CATGGCTCGTTCCTTACGCCGACATCCTTACTCTTCTCCTGGCATTGTTTATT
LipB	ATGAAAAAAGTACTTATGGCATTTCATTATTTGTTTATCGCTGATTCTATCTGT TTAGCCGCTCCGCCGCTCTGGCGCAAAGCT
LipA	ATGAAATTTGTAAGAAGAAGGATCATTGCACTTGTAACAATTTTGATGCTGT CTGTTACATCGCTGTTTGCCTTGACGCCGTCAGCAAAGCC

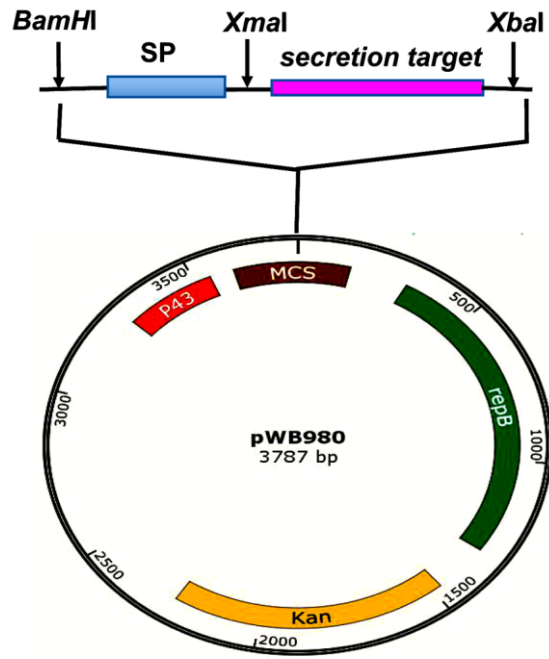


Fig. S1 Construction of plasmids carrying different signal peptides.

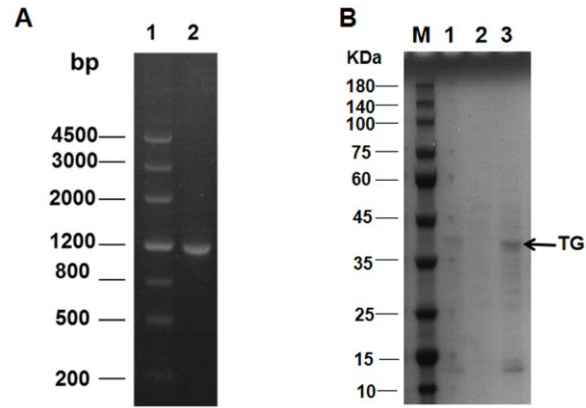


Fig. S2 PCR amplification of the encoding gene of *S. ladakanum* TG (A). Lane 1, Marker; Lane 2, encoding gene of *S. ladakanum* TG. SDS-PAGE analysis of *S. ladakanum* TG (B). M, Marker; Lane 1, intracellular fraction of *B. subtilis* WB600 carrying plasmid pWB_{sacB-LTG}; Lane 2, extracellular fraction of *B. subtilis* WB600 carrying plasmid pWB_{sacB}.; Lane3, extracellular fraction of *B. subtilis* WB600 carrying plasmid pWB_{sacB-LTG}.