

Supplementary data

Figure S1. Growth profiles of the recombinant *L. plantarum* WCFS1 strains harboring AmyL (A) and AmyA (B) secretion plasmids with different signal peptides in 200 mL of MRS containing 5 μ g/mL erythromycin at 37°C. Values given are the average values from at least two independent experiments and the error bars indicate the standard deviation.

Figure S2. SDS-PAGE analysis of cell-free supernatant and lysate of *L. plantarum* WCFS1 strains at 9 h after induction overexpressing the ~100 kDa α -amylase encoded by *amyL* (A) and the ~50 kDa α -amylase encoded by *amyA* (B). The arrows indicate the bands of α -amylases, M denotes the Precision protein ladder (Bio-Rad). Non-induced strains harbouring the plasmid pLp_spAmyA_AmyL and pLp_spAmyA_AmyA were representatives for non-induced conditions in (A) and (B), respectively.

Tables

Table S1. Primers used for cloning in this study

Primers	Primer sequences (5'→3')	Restriction site underlined
AmyL_SalI_Fw	GGCGGAGTCGACGATAGTTATACGACAT CAACTGATGAC	<i>SalI</i>
AmyL_EcoRI_Rv	AGTAGTGAATTCTTACGAAGTGCTTGATG TGC	<i>EcoRI</i>
401_spAmyA_EcoRI_Fw	GATAAGAATTCGGTACCCCGGGTTCGAA	<i>EcoRI</i>
401_spAmyA_SalI_Rv	GATTAGTCGACACTAGCCGCTTGAGCAA CTTGTTTAGA	<i>SalI</i>

Table S2. Strains and plasmids used in this study

Strains and plasmids	Characteristics	References/Sources
Strains		
<i>E. coli</i> NEB5 α	Host strain	New England Biolabs (Frankfurt am Main, Germany)
<i>L. plantarum</i> WCFS1	Host, protein expression strain	Kleerebezem <i>et al.</i> (2003)
<i>L. plantarum</i> S21	Native strain	Kanpiengjai <i>et al.</i> (2014)
Plasmids		
pSIP401	<i>spp</i> -based expression vector with inducible promoter P _{sppA} ; Erm ^R	Sørvig <i>et al.</i> (2003)
pSIP409	<i>spp</i> -based expression vector with inducible promoter P _{sppQ} , Erm ^R	Sørvig <i>et al.</i> (2005)
<i>pSIP401 derivatives</i>		
pLp_2145s_AmyA	<i>amyA</i> fused to Lp_2145	Mathiesen <i>et al.</i> (2009)
pLp_3050s_AmyA	<i>amyA</i> fused to Lp_3050	Mathiesen <i>et al.</i> (2008)
pLp_0373s_AmyA	<i>amyA</i> fused to Lp_0373	Mathiesen <i>et al.</i> (2008)
pLp_spAmyA_AmyA	<i>amyA</i> with its native signal peptide (SP_AmyA)	Mathiesen <i>et al.</i> (2008)
pLp_2145s_AmyL	<i>amyL</i> fused to Lp_2145	This study
pLp_3050s_AmyL	<i>amyL</i> fused to Lp_3050	This study
pLp_0373s_AmyL	<i>amyL</i> fused to Lp_0373	This study
pLp_spAmyA_AmyL	<i>amyL</i> fused to signal peptide of AmyA (SP_AmyA)	This study
<i>pSIP409 derivative</i>		
pLp_AmyL7	<i>amyL</i> with native signal peptide (SP_AmyL)	Kanpiengjai <i>et al.</i> (2015b)

Table S3. Sequences of the signal peptides used in this study.

Signal peptides	Amino acid sequences of the signal peptides (including 2 amino acids downstream of cleavage site) ^a
SP_AmyA	MKKKKSFWLVSFLVIVASVFFISFGFSNHSKQVAQA↓AS
SP_AmyL	MKKKKSFWLVSFLVIVASVFFISFGLSNHSNQVAQA↓DS
Lp_2145	MKKINKLMILGMLVFGVTGATMINPEMTTAAHA↓SA
Lp_3050	MKKFNFKTMLLLVLASCVFGVVVNVTSLGPQTAITAQA↓SK
Lp_0373	MYTENTGKHHRNGLPVWLLPLLVVISFWGVSQNIMVVDA↓SS

^aThe arrows ↓ represent the cleave sites of the signal peptides as determined by SignalP-5.0 (www.cbs.dtu.dk/services/SignalP/) (Armenteros *et al.*, 2019)