

Supplementary Material

Identification of Marker Peptides for the Whey Protein Quantification in Edam-Type Cheese

Tobias von Oesen, Mascha Treblin, Ingrid Clawin-Rädecker, Dierk Martin, Ronald Maul, Wolfgang Hoffmann, Katrin Schrader, Benjamin Wegner, Katja Bode, Ralf Zink, Sascha Rohn, and Jan Fritsche

Contents

Table S1	Overview of the volumes used for peptide spiking in duplicate and measured once.
Table S2	Preparation of the synthetic peptide standard mixture (according to the peptide spiking levels mentioned in Table S1) in duplicate and measured once.
Table S3	Spiking of CPBS with MPSM prior to tryptic hydrolysis.
Table S4	Overview of the sample volumes used for the tryptic hydrolyses of the spiking experiments (Table S3).
Figure S1	Peak areas of tryptic <i>in-vitro</i> generated PMPs for α -LA or β -LG throughout the cheese ripening process of sample set I.
Figure S2	Peak areas of tryptic <i>in-vitro</i> generated PMPs for α -LA or β -LG throughout the cheese ripening process of sample set II.
Figure S3	Peak areas of PMPs as a function of the amount of HH milk in cheese milk for sample set I and sample set II.

Table S1. Overview of the volumes used for peptide spiking in duplicate and measured once.

Sample	Hydrolysate [μL]	Added synthetic peptide standard mixture [μL]	Water [μL]
Hydrolysates of CPBS and MPSM	300	0	300
Peptide spiking level 1	300	150	150
Peptide spiking level 2	300	225	75
Peptide spiking level 3	300	300	0

Table S2. Preparation of the synthetic peptide standard mixture (according to the peptide spiking levels mentioned in Table S1) in duplicate and measured once.

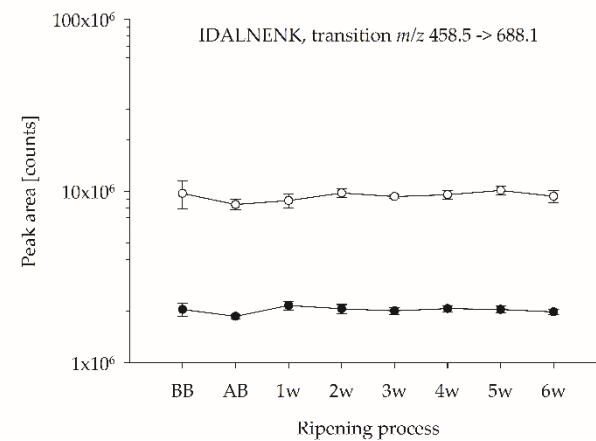
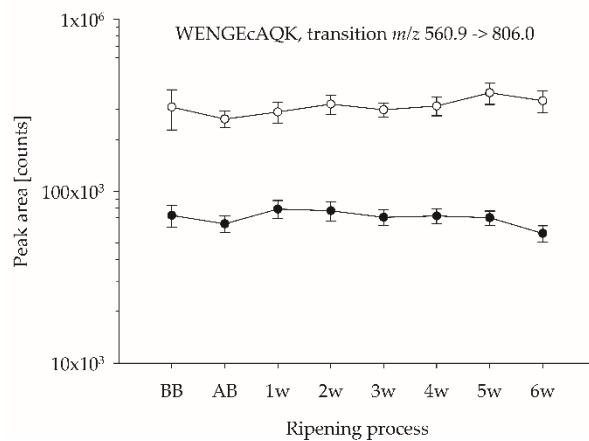
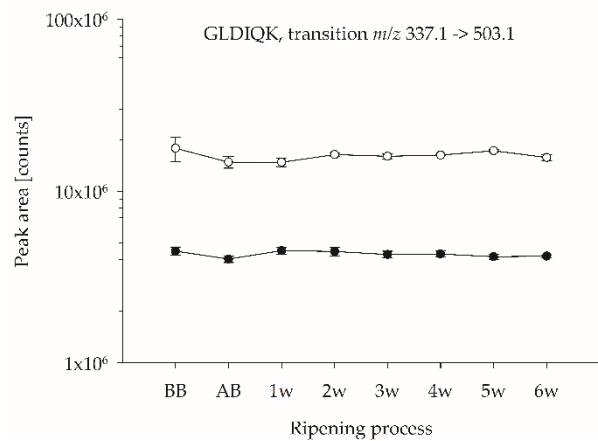
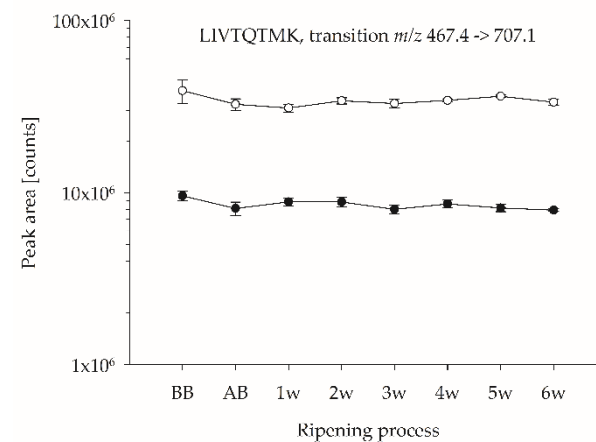
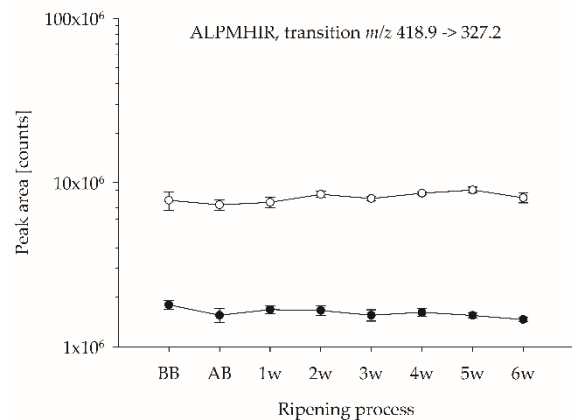
Peptide spiking level	Synthetic peptide standard mixture [μL]	Water [μL]
1	150	450
2	225	375
3	300	300

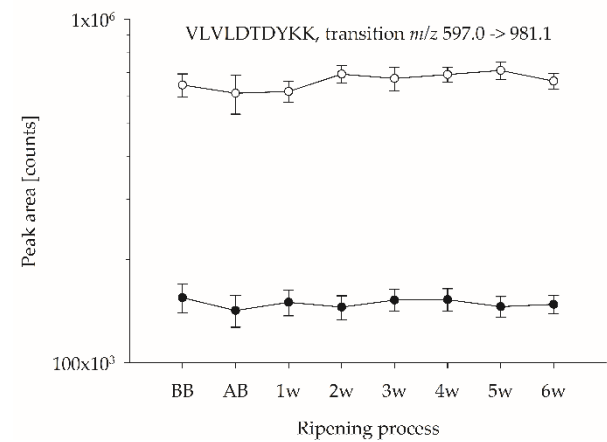
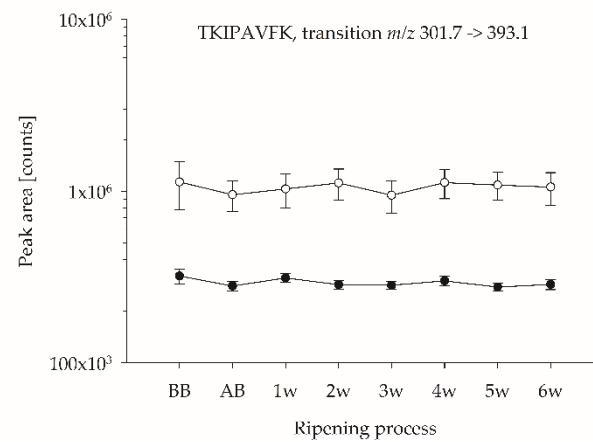
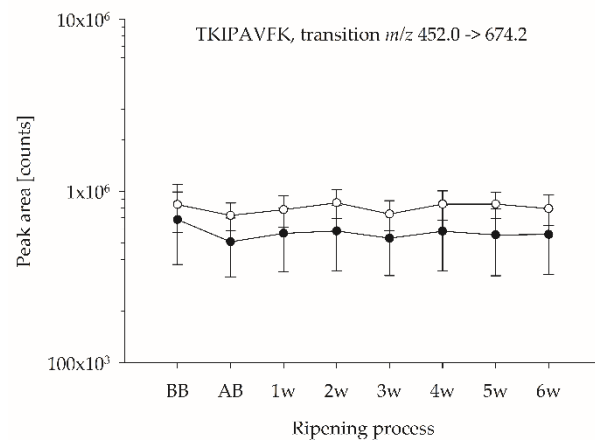
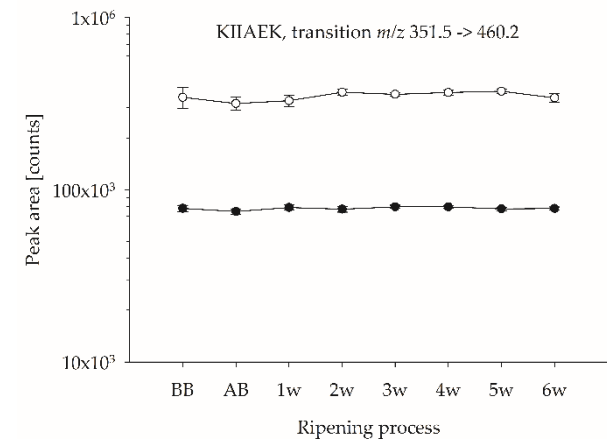
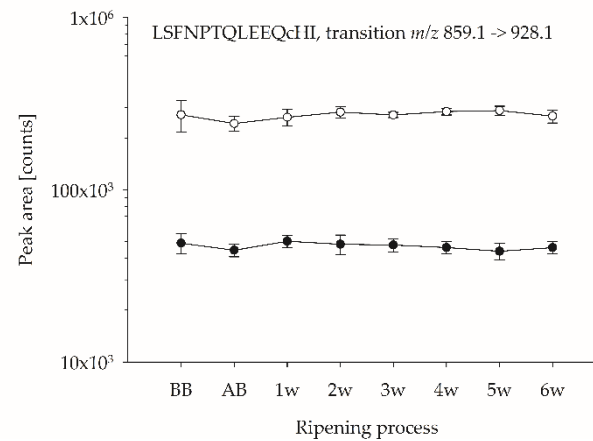
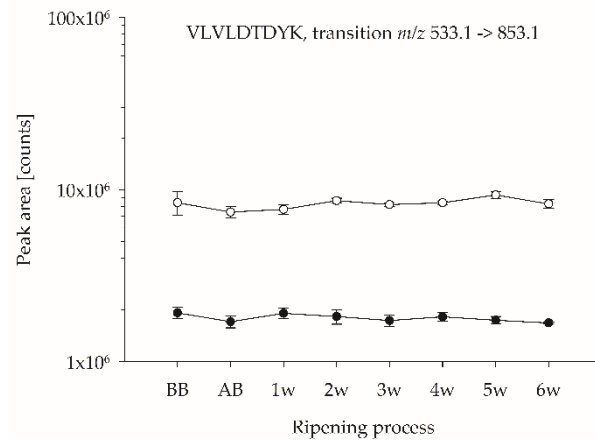
Table S3. Spiking of CPBS with MPSM.

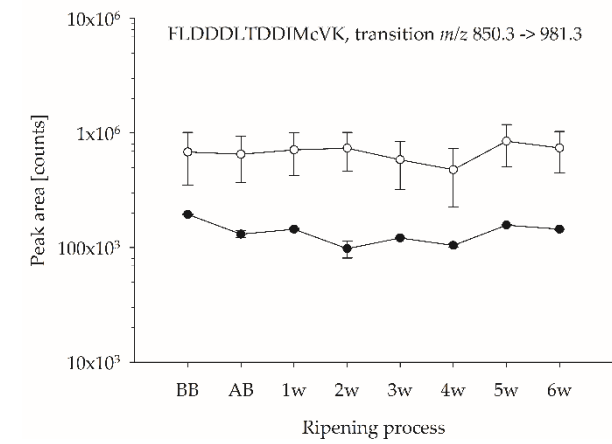
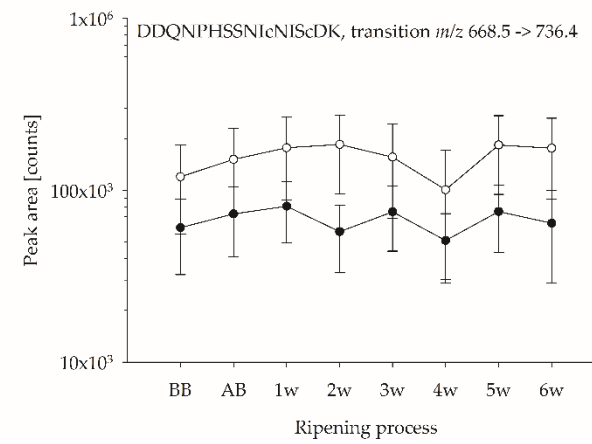
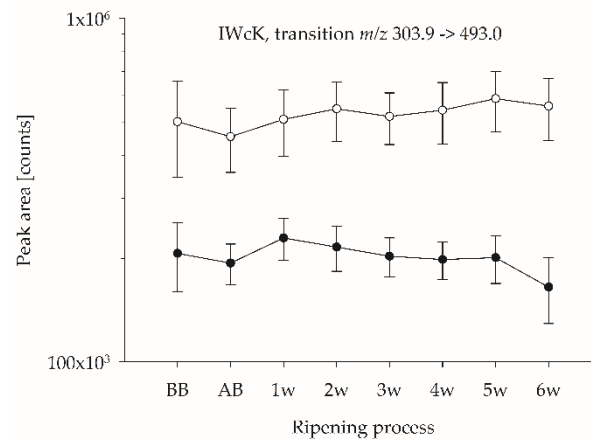
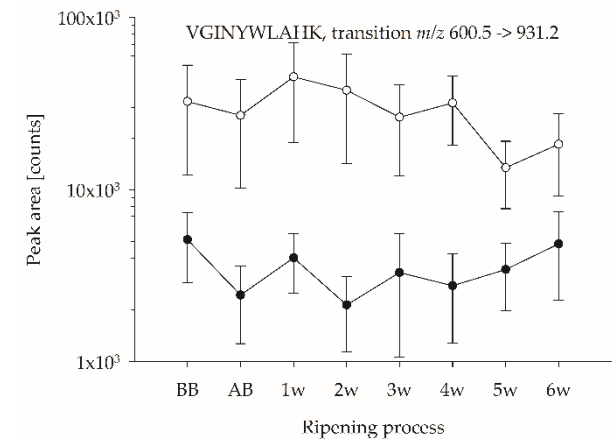
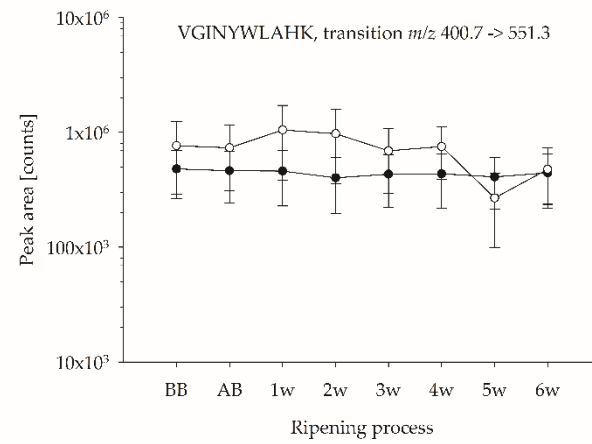
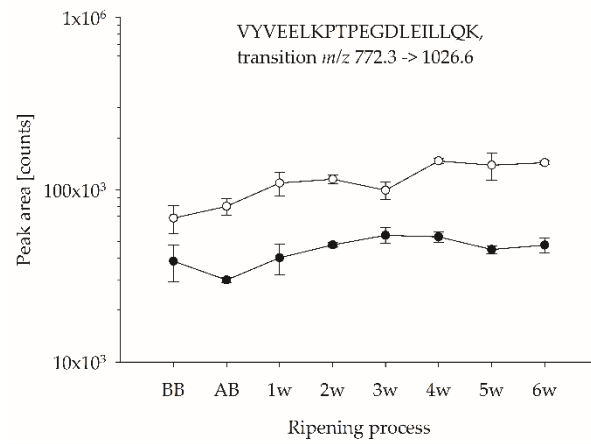
Spiking level	CPBS [μL]	MPSM [μL]	Water [μL]
1	50	50	100
2	50	100	50
3	50	150	0

Table S4. Overview of the sample volumes used for tryptic hydrolysis.

Sample	Volume used for hydrolysis [μL]	Volume of CPBS present in sample volume of the spiked samples [μL]	Volume of MPSM present in sample volume of the spiked samples [μL]	Water [μL]
CPBS	20	–	–	700
MPSM level 1	20	–	–	700
MPSM level 2	40	–	–	680
MPSM level 3	60	–	–	660
Spiking level 1	40	10	10	680
Spiking level 2	40	10	20	680
Spiking level 3	40	10	30	680







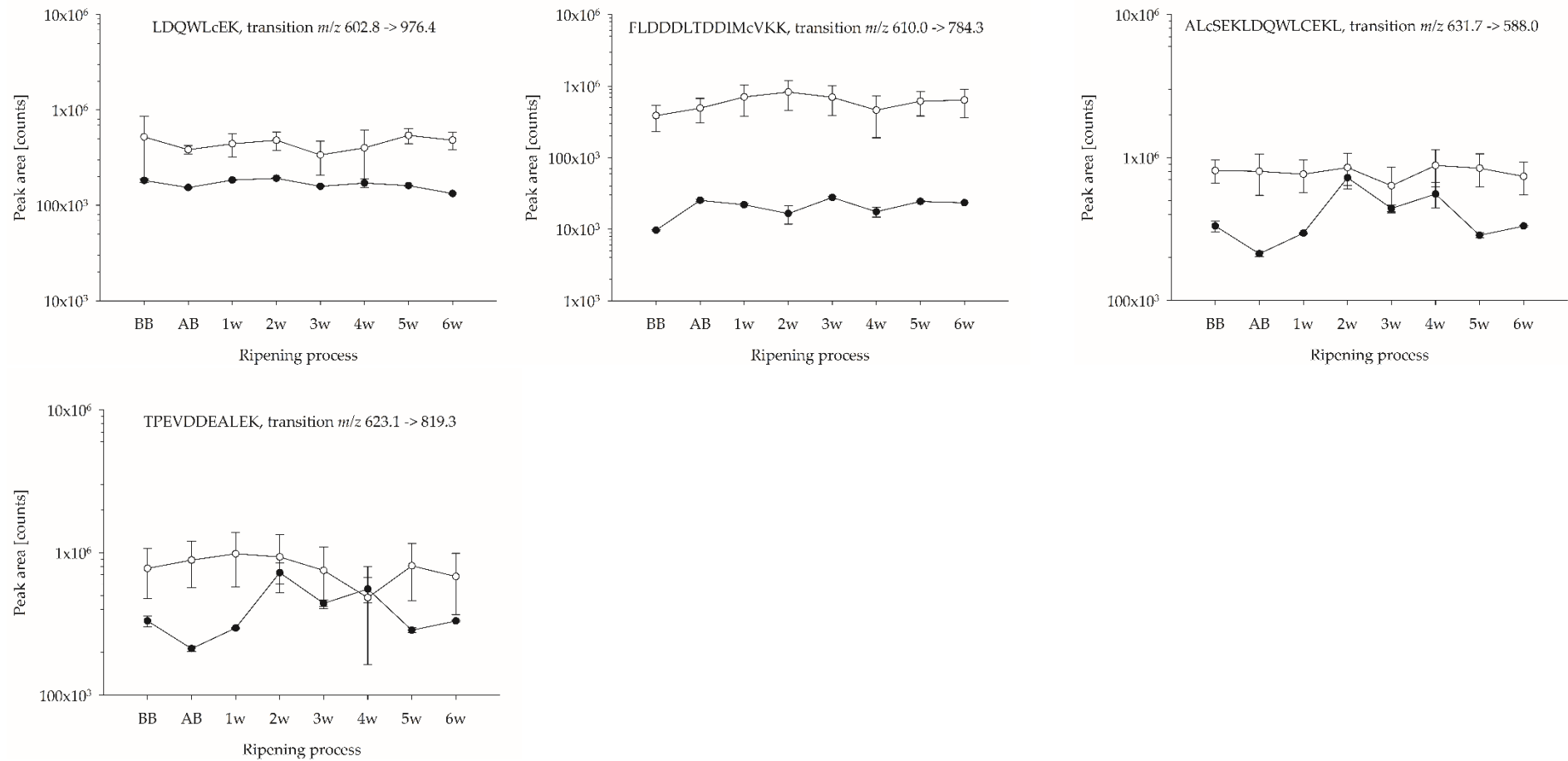
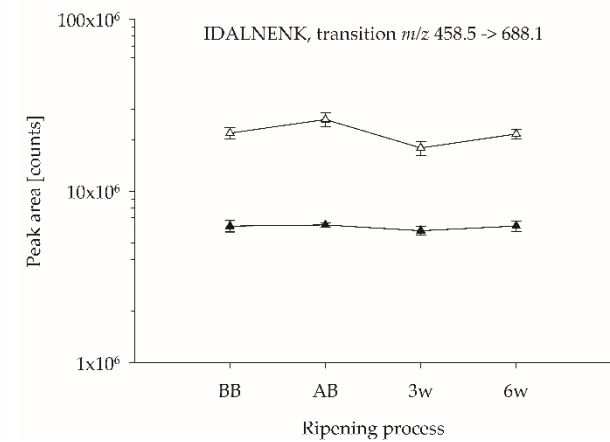
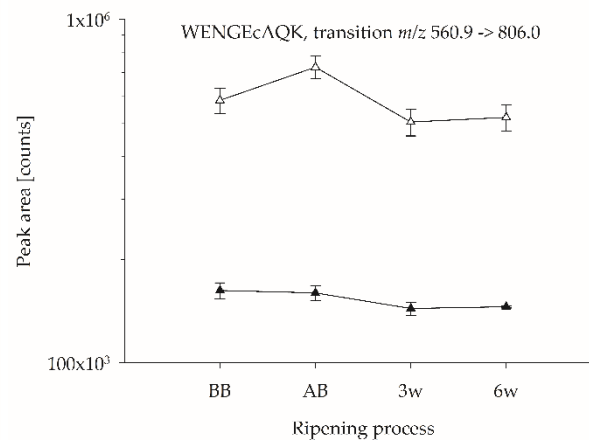
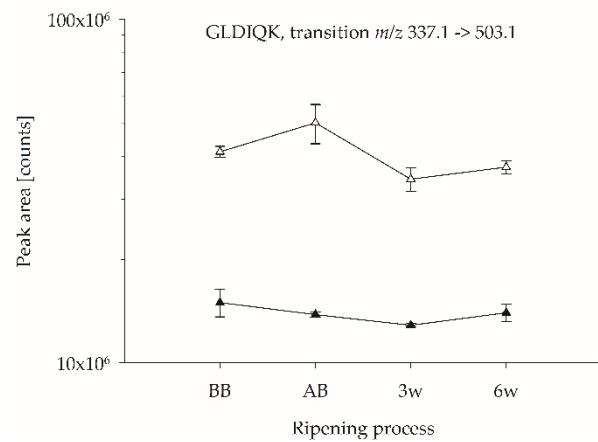
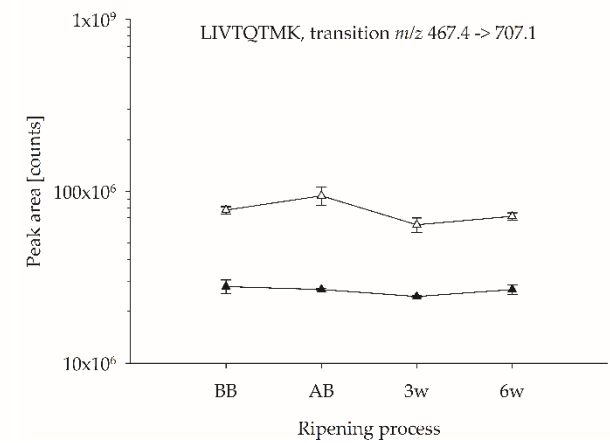
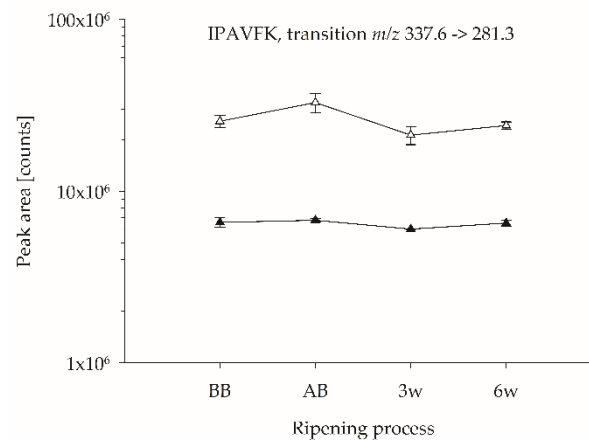
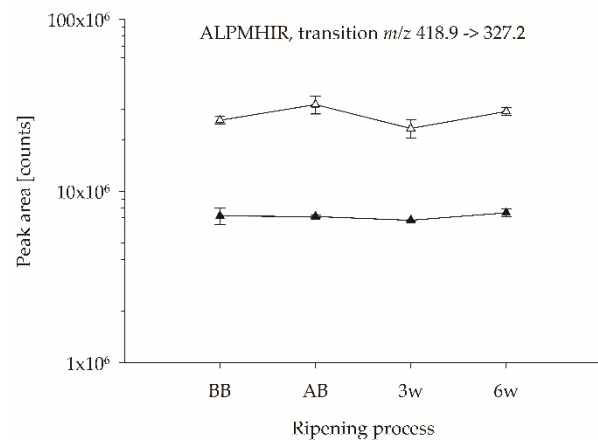
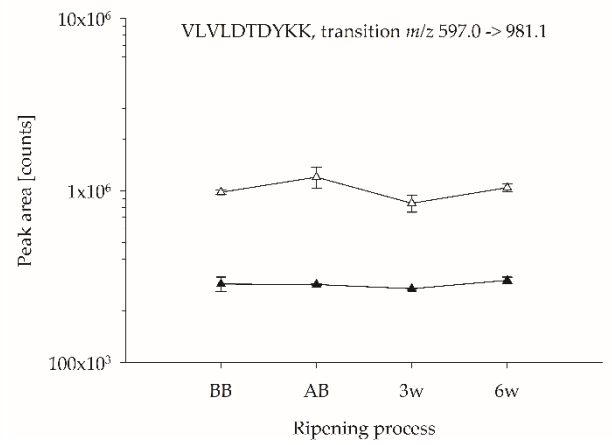
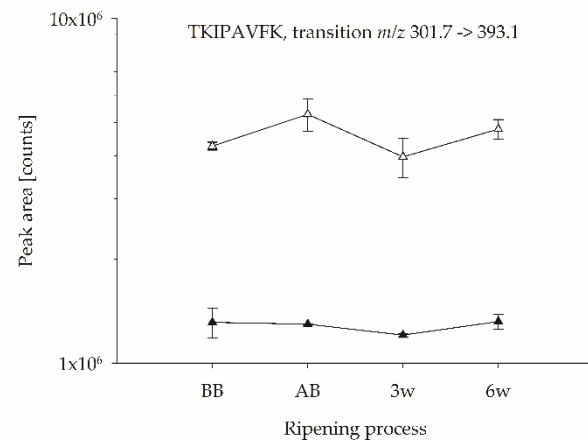
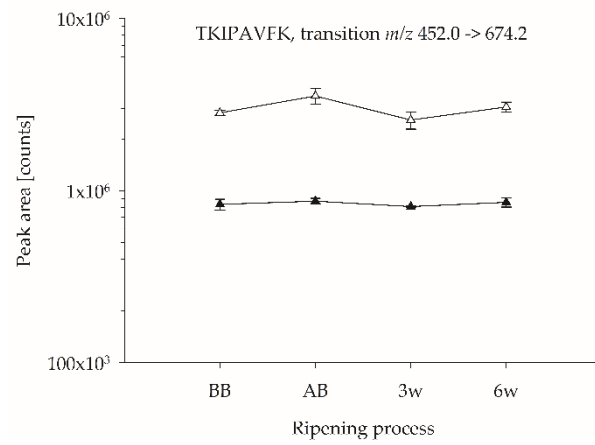
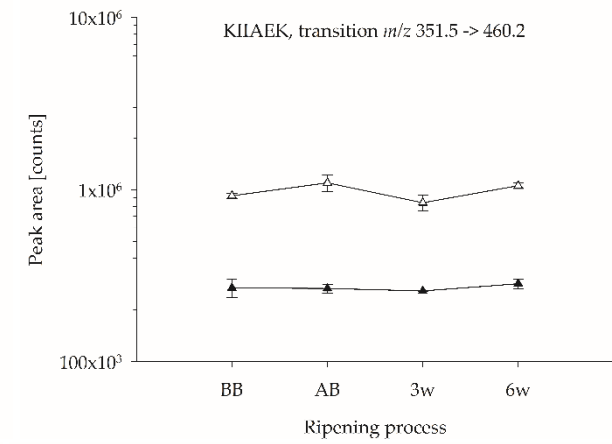
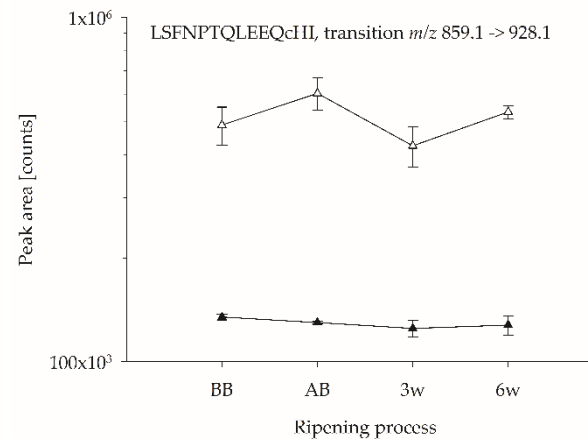
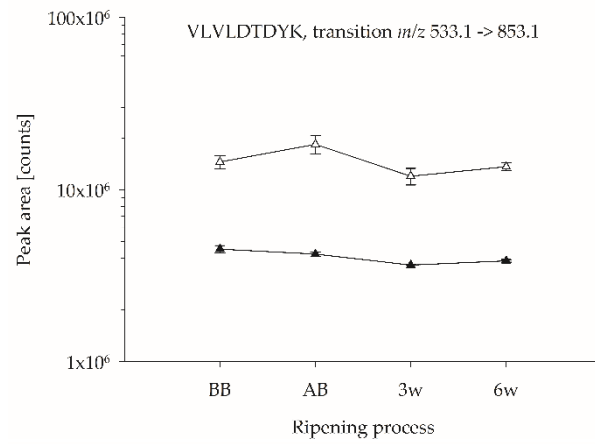
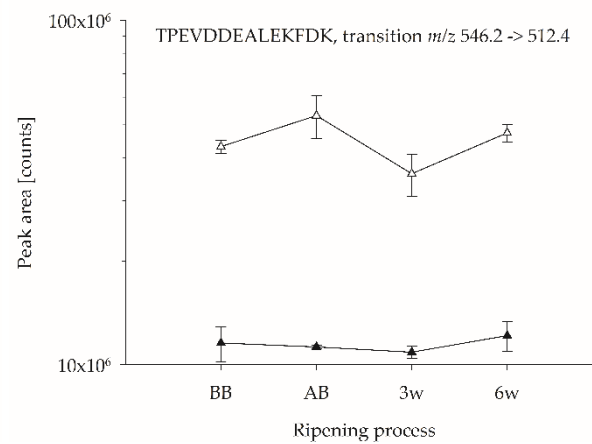
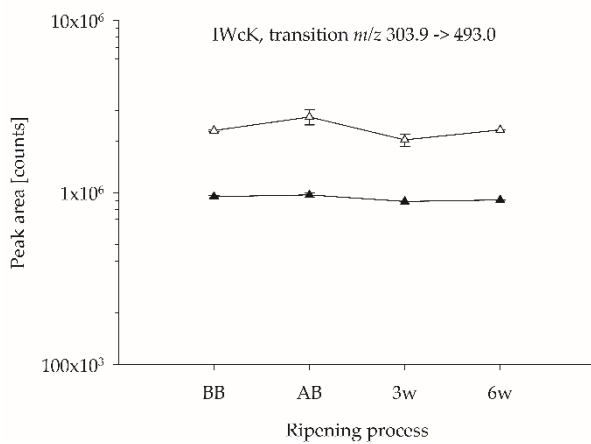
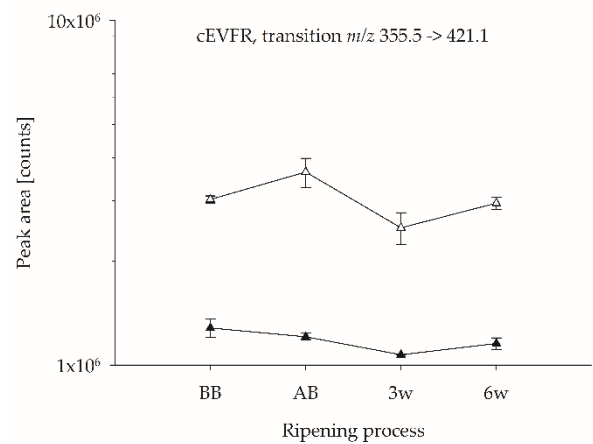
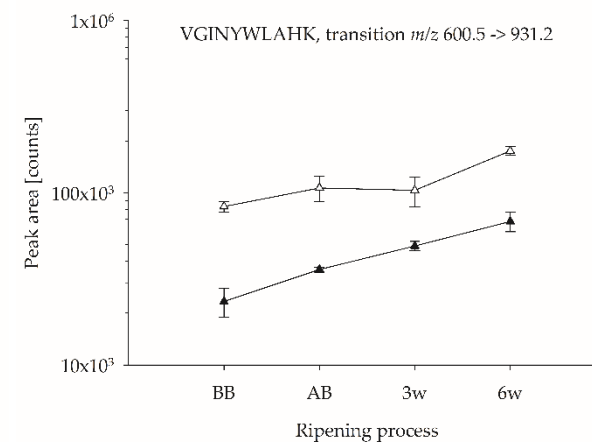
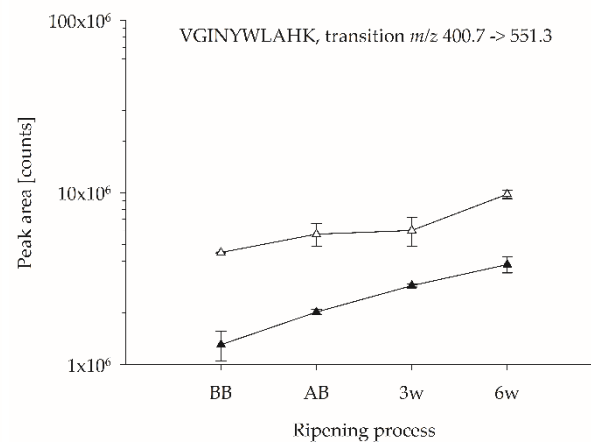
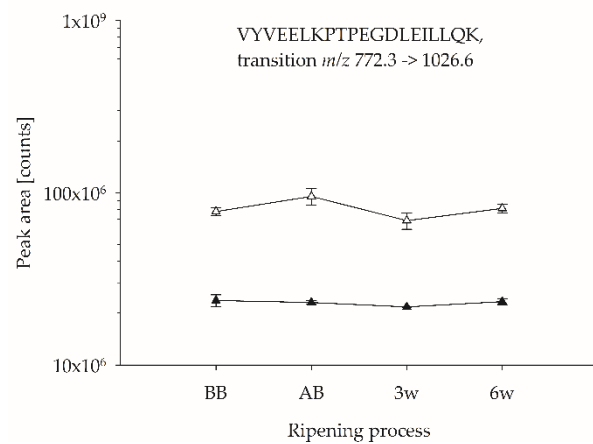


Figure S1. Peak areas of tryptic *in-vitro* generated PMPs for α -LA or β -LG throughout the cheese ripening process of sample set I. For graphical representation, the y-axis was displayed logarithmically. Data were calculated as mean \pm SD of three replicate productions; (●) cheese containing 0% HH milk; (○) cheese containing 30% HH milk. Sample preparation was carried out in duplicate and measured once.







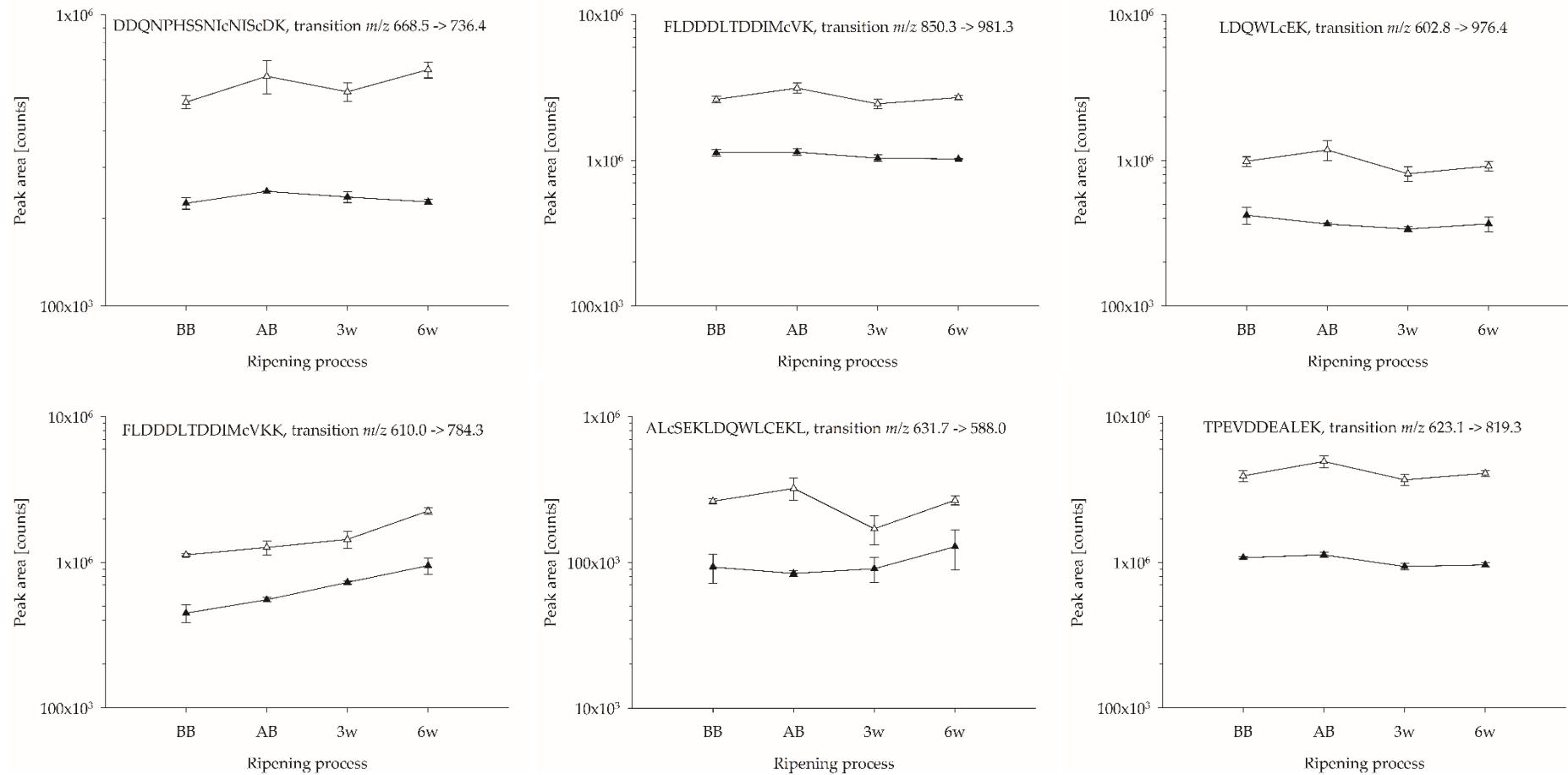
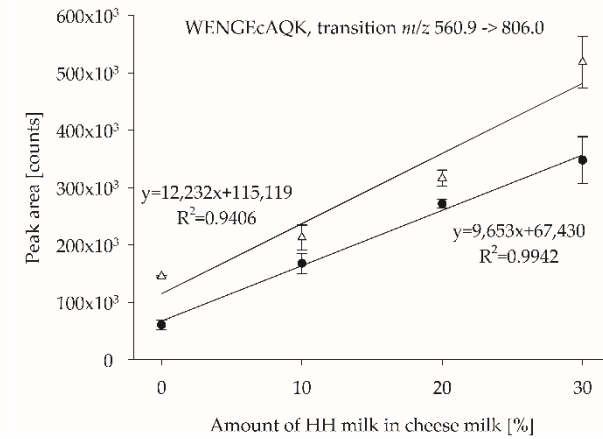
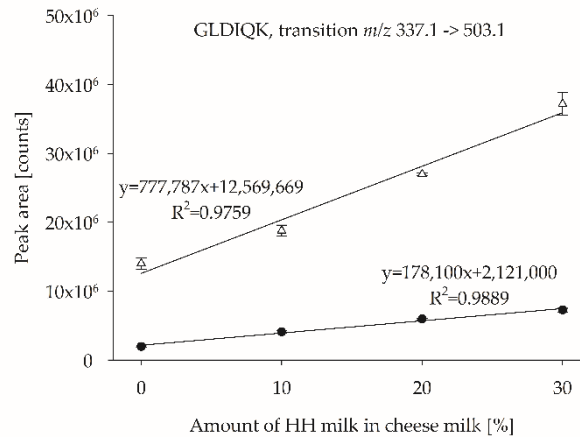
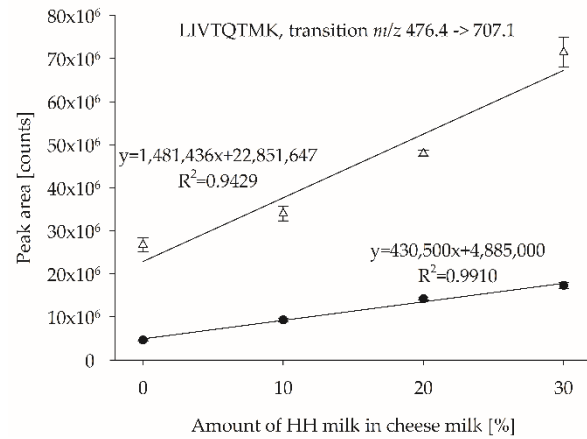
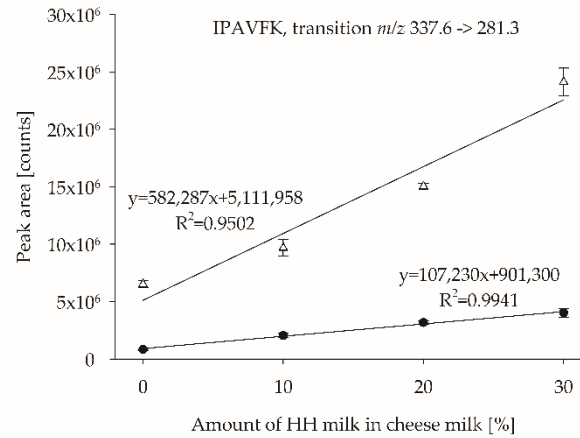
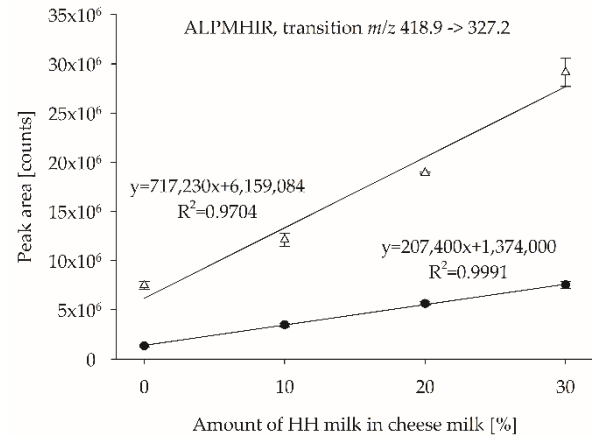
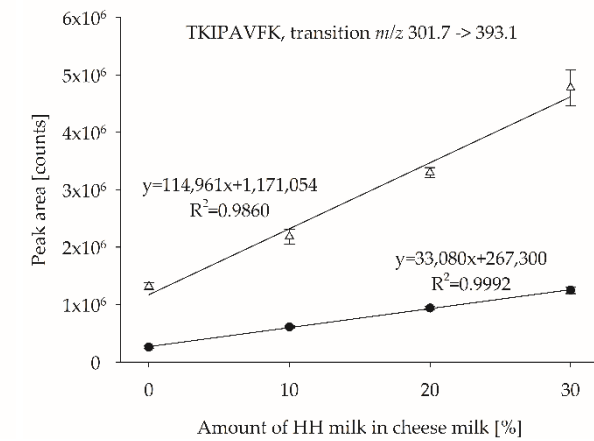
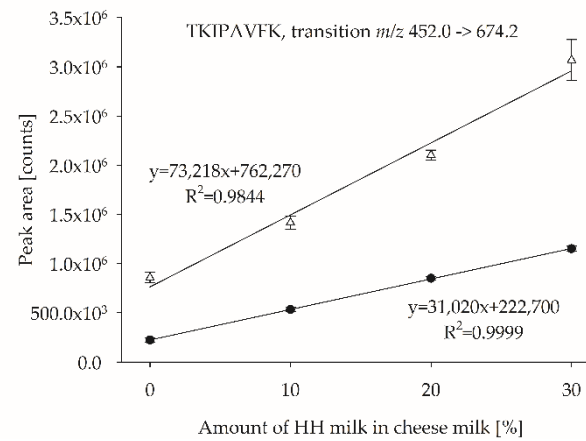
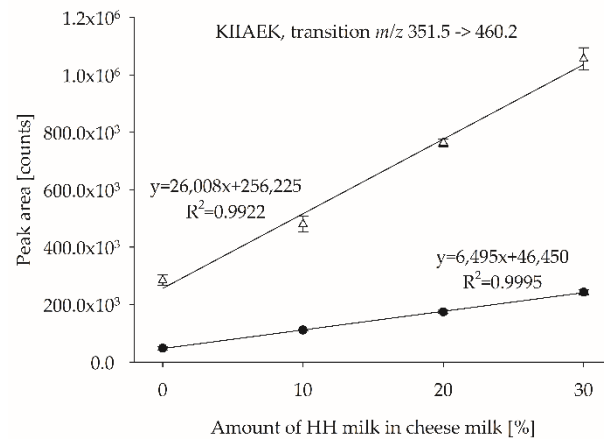
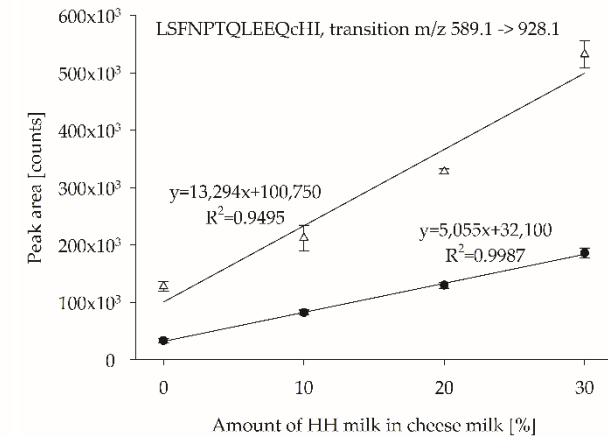
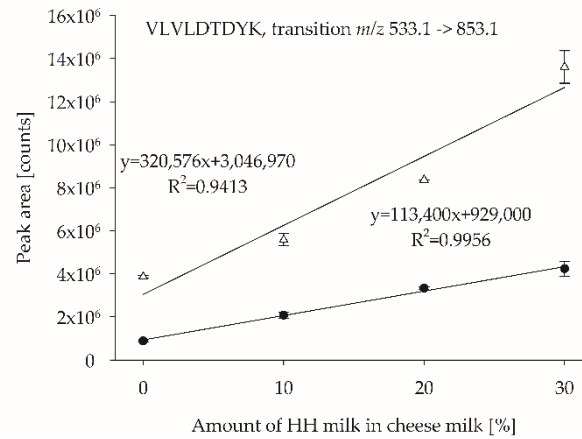
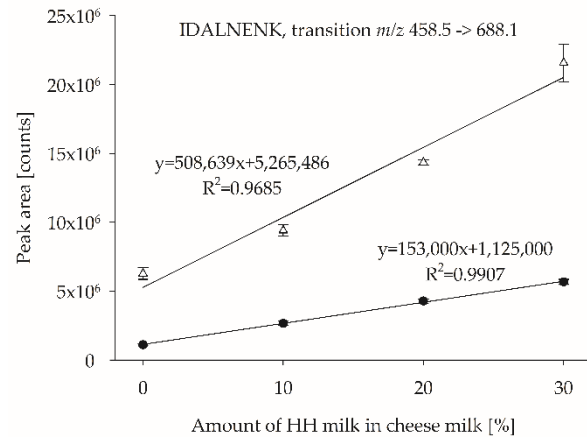
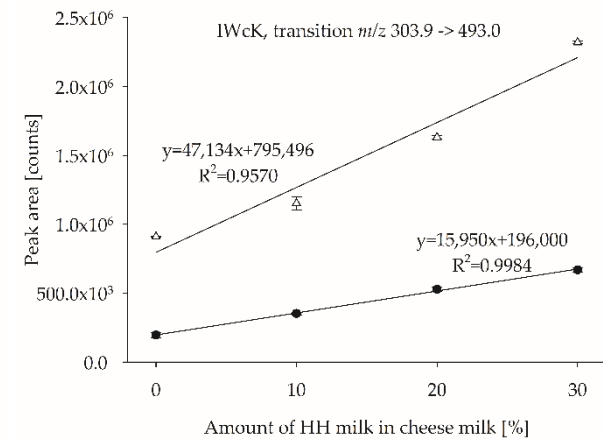
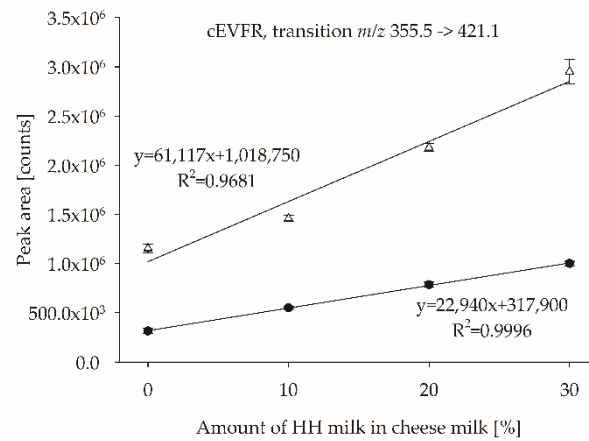
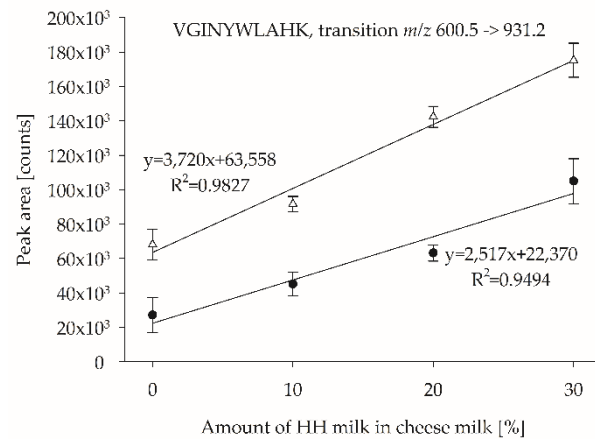
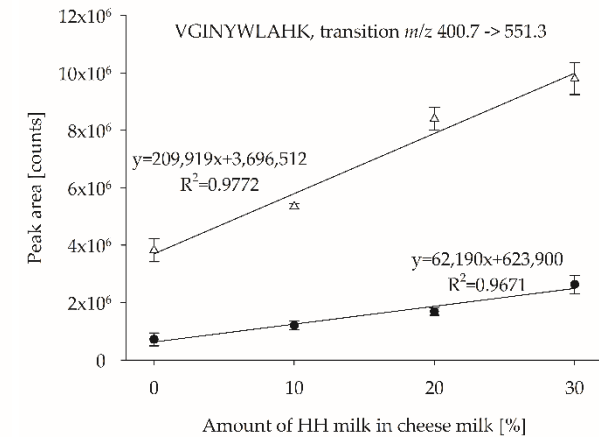
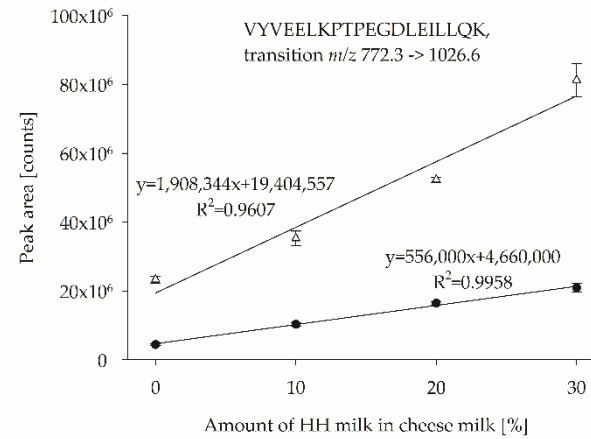
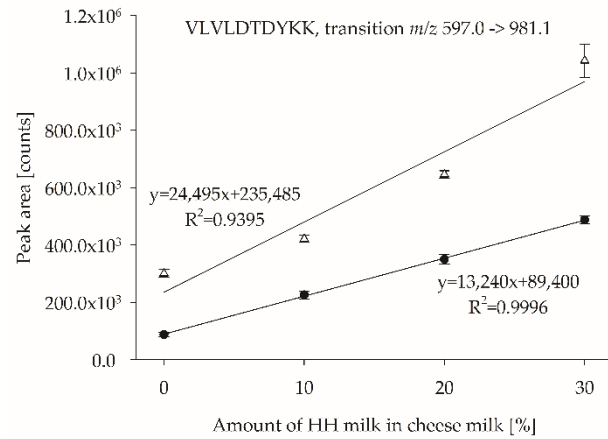
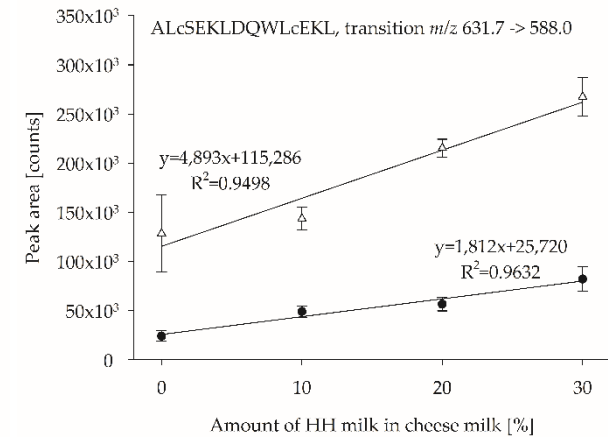
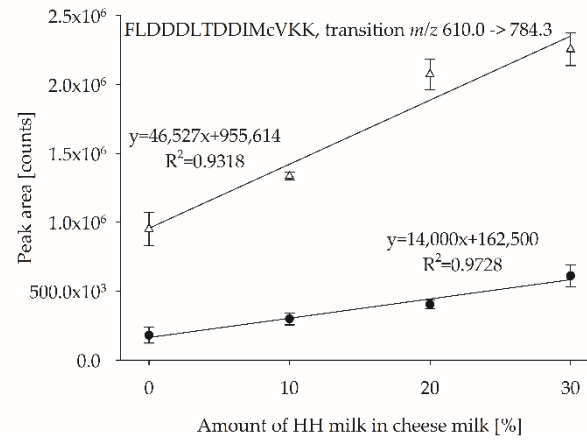
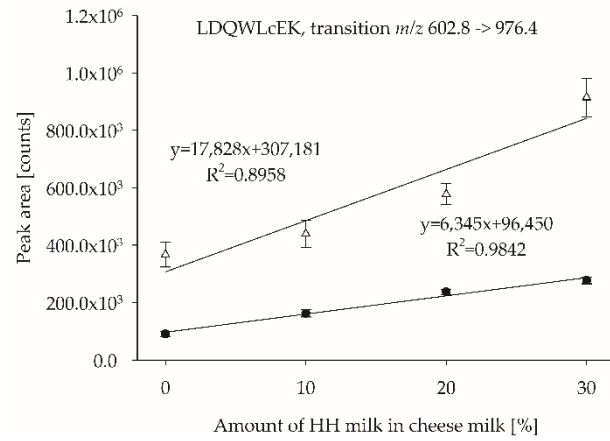
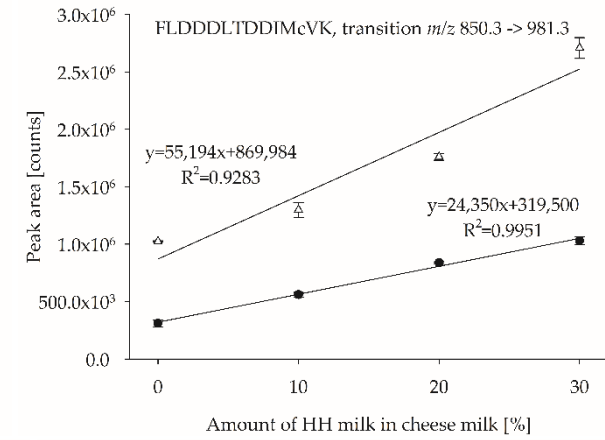
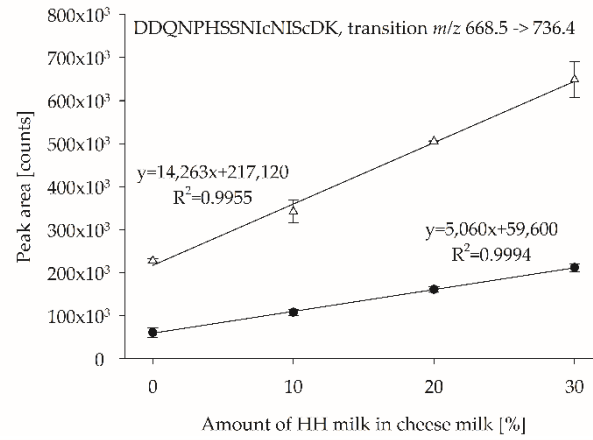
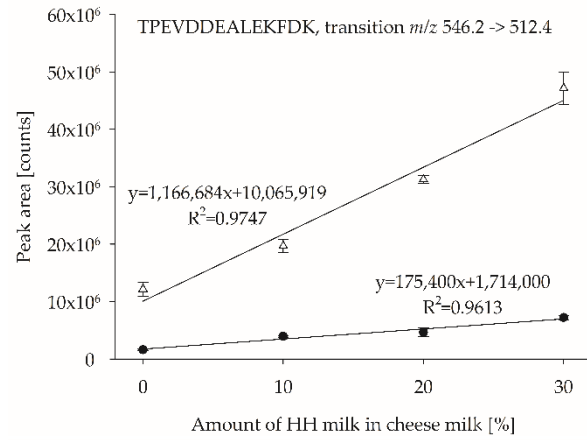


Figure S2. Peak areas of tryptic *in-vitro* generated PMPs for α -LA or β -LG throughout the cheese ripening process of sample set II. For graphical representation, the y-axis was displayed logarithmically. Data were calculated as mean \pm SD of a sample preparation in duplicate and measured once; (\blacktriangle) cheese containing 0% HH milk, (\triangle) cheese containing 30% HH milk.









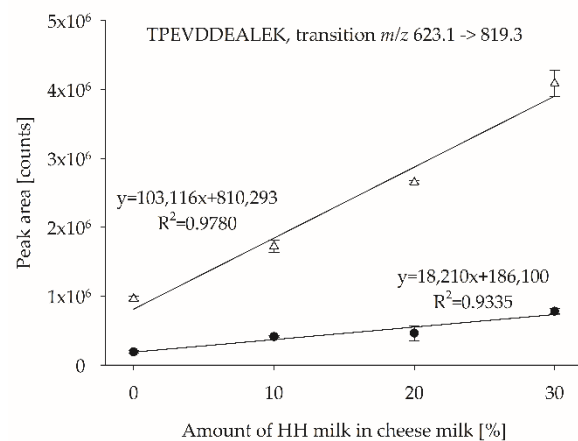


Figure S3. Peak areas of PMPs as a function of the amount of HH milk in cheese milk for sample set I (\bullet) and sample set II (Δ). R^2 stands for the coefficient of determination. Data points were calculated as mean \pm SD. Sample preparation was carried out in duplicate and measured once.