SUPPLEMENTARY MATERIAL

Identification of emulsifier potato peptides by bioinformatics: application to omega-3 delivery emulsions and release from potato industry side streams

Pedro J. García-Moreno[1*], Simon Gregersen[2], Elham R. Nedamani[1], Tobias H. Olsen[3], Paolo Marcatili[3], Michael T. Overgaard[2], Mogens L. Andersen[4], Egon B. Hansen[1], Charlotte Jacobsen[1]

[1] National Food Institute, Technical University of Denmark, Denmark
[2] Department of Chemistry and Bioscience, Aalborg University, Denmark
[3] Department of Bio and Health Informatics, Technical University of Denmark, Denmark
[4] Department of Food Science, University of Copenhagen, Denmark

Pedro J. García-Moreno and Simon Gregersen contributed equally to this work.

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[*] Corresponding author: pejeg@food.dtu.dk

Table 1S. Initially selected proteins from *Solanum tuberosum* used to identify emulsifier peptides by bionformatics

Entry	**Protein**	**Length**
P15478	Patatin-T5	386
P15477	Patatin-B2	386
Q3YJT5	Patatin-05	387
P11768	Patatin group M-1	386
Q2MY60	Patatin-02	387
Q2MY43	Patatin-08	387
Q42502	Patatin-2-Kuras 3	386
Q3YJS9	Probable inactive patatin-3-Kuras 1	374
Q3YJT3	Patatin-2-Kuras 1	374
Q8RXA3	Cathepsin D inhibitor (KTI-A protein) (Kunitz-type enzyme inhibitor P4E1)	220
Q3S474	KTI-B protein (Kunitz-type protease inhibitor)	221
Q3S477	KTI-B protein (Kunitz-type protease inhibitor)	219
Q2XPX8	KTI-B protein (Kunitz-type protease inhibitor-like protein)	213
Q3S488	Kunitz-type protease inhibitor	220
Q3S481	Kunitz-type protease inhibitor	220
A0A097H149	Kunitz-type inhibitor B	221
Q38M77	Kunitz-type protease inhibitor	220
Q3S489	Kunitz-type protease inhibitor	221
Q3S482	Kunitz-type protease inhibitor	220
Q3S479	Kunitz-type protease inhibitor	220
Q8H9D6	Kunitz-type trypsin inhibitor	220
Q2XPY0	Kunitz-type protease inhibitor-like protein	213
M1LA62	Kunitz-type protease inhibitor D	219
Q8W126	Kunitz-type enzyme inhibitor S9C11	242
L0AQL9	Kunitz-type proteinase inhibitor PKI2	221
A0A097H120	Clone PI8311 Kunitz-type inhibitor A/B mRNA	203
Q66LL8	Kunitz-type proteinase inhibitor	194
Q66LL2	Kunitz-type proteinase inhibitor group B	194
H9B8J1	Kunitz-type proteinase inhibitor group A1	191
H9B8I9	Kunitz-type proteinase inhibitor group A1	191
Q8LJQ0	Kunitz-type proteinase inhibitor	192
H9B8I8	Kunitz-type proteinase inhibitor group A1	191
H9B8J0	Kunitz-type proteinase inhibitor group A1	190
M1KPH5	Kunitz-type protease inhibitor C	178

Table 2S. Detailed description of the ramped gradient used for LC-MS/MS analysis of potato side stream samples.

Time [mm:ss]	Duration [mm:ss]	Mixture [%B]	Flow [nL/min]
00:00	00:00	5	300
01:00	01:00	5	300
05:00	04:00	12	300
145:00	140:00	37	300
150:00	05:00	50	300
151:00	01:00	100	300
165:00	14:00	100	300
170:00	05:00	5	300

Table 3S: Summary statistics for LC-MS/MS proteomics analysis of potato side stream samples using MaxQuant.

Sample	**AKV-Feed**	**AKV-K2**	**KMC-Feed**	**KMC-Food**	**AKV-PFJ**
# total peptides (matched)	1229	1034	1208	3958	7840
# total protein groups	394	334	342	1007	1689
# contaminant proteins	24	26	52	31	41
# reverse protein hits	3	4	3	11	24
# potato protein groups	367	304	287	965	1624
# potential potato proteins	1154	948	912	2356	3633

Table 4S. Identified and quantified patatin isoforms by LC-MS/MS. Proteins were identified by their UniProt AC# and quantified sample-wise using the iBAQ algorithm in MaxQuant (normalized to riBAQ).

AKV-Feed		**AKV-K2**		**KMC-Feed**		**KMC-Food**		**AKV-PFJ**	
AC#	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**
Q3YJT3	22.6%	Q3YJT3	13.7%	P15477	29.6%	Q2MY60	11.2%	Q2MY45	12.7%
Q2MY44	20.2%	Q2MY44	9.3%	Q2MY44	12.5%	Q2MY59		Q2MY36	
Q3YJS9	4.0%	Q3YJS9	1.4%	Q3YJS9	4.9%	Q2MY58		P15476	
Q2VBJ3		Q2VBJ3		Q2VBJ3		Q2MY50		Q2MY42	
Q2VBJ2		Q2VBJ2		Q2VBJ2		Q3YJT4		Q3YJS9	4.8%
Q2MYJ6		Q2MYJ6		Q2MYJ6		Q41487		Q2VBJ3	
Q2MYJ4		Q2MYJ4		Q2MYJ4		Q2MYW0		Q2VBJ2	
Q2MYC5		Q2MYC5		Q2MYC5		Q2MYK2		Q2MYJ6	
Q2MY85		Q2MY85		Q2MY85		Q2VBI1		Q2MYJ4	
Q2MY98		Q2MY88		Q2MYB7		Q2MY36	9.3%	Q2MYC5	
Q2MYF1				Q2MY88		Q2MY45		Q2MY85	
Q2MYE9				Q9AUH5		Q2MY42		Q2MY88	
Q2MYB8				M1BFJ1		P15476		Q2MYB7	
Q2MY88				Q3YJT3	4.0%	Q3YJS9	2.7%	Q2MY44	3.2%
Q2MYB7				Q3YJT4	3.8%	Q3YJT3	1.8%	Q3YJT3	1.3%
Q3YJT4	3.8%			Q2MY60				Q3YJT4	1.2%
Q2MY60				Q2MY59				Q2MY60	
Q2MY59				Q2MY58				Q2MY59	
Q2MY58				Q2MY50				Q2MY58	
Q2MY50				Q41487				Q2MY50	
Q2VBJ0				Q2VBJ0				Q2MYW0	
Q2VBI9				Q2VBI9				Q2MYK2	
Q2VBI6				Q2VBI6					
Q2MYY2				Q2MYX4					
Q2MYX5				Q2MYW0					
Q2MYX4				Q2MYV6					
Q2MYW0				Q2MYV1					
Q2MYV6				Q2MYT1					
Q2MYV1				Q2MYS7					
Q2MYT1				Q2MYS6					
Q2MYS9				Q2MYS2					
Q2MYS7				Q2MYQ3					
Q2MYS6				Q2MYN1					
Q2MYS2				Q2MYK2					
Q2MYQ3				Q2MYJ9					
Q2MYN1				Q2MYG9					
Q2MYK2				Q2MYF3					
Q2MYJ9				Q2MYE1					
Q2MYJ1				Q2MY96					
Q2MYH4				Q2MYM3					
Q2MYG9				Q2MYH4					

Q2MYF4 Q2MYF3 Q2MYE1 Q2MY96 Q2MYM3 Q2MYW8 Q2MY72 Q2VBI7 Q2MYU1 Q2MYM8 Q2MY67 Q2MYW1 Q7DMV4	
Q3YJT0	1.6%
Q2VBI2 Q2MY99	1.1%
Q3YJT2	1.0%

Q2MYF4 Q2MYX7 Q2MYW4 Q2MYU3 Q2MYS5 Q2MYP2 Q2MYK7 Q2MYE8 Q2MYW8 Q2MY72 Q2VBI7 Q2MYU1 Q2MYM8 Q2MY67 Q2MYW1	
Q42502	1.3%
P07745 M1BFJ0	1.1%

Table 5S. Identified and quantified protease inhibitors (KTIs, PINs, and MCPIs) by LC-MS/MS. Proteins were identified by their UniProt AC# and quantified sample-wise using the iBAQ algorithm in MaxQuant (normalized to riBAQ).

AKV-Feed	
AC#	**riBAQ**
Q3S488 Q3S482 Q3S479	8.8%
Q41484 Q2XPX8 P58517	5.1%
O24385 Q8RX93 A0A097H166 O24386 Q8RX92 A0A097H155	3.7%
P58515 Q8L5X4 M1AKE4 Q66LL8 Q66LL2 Q8L5X3 Q8L5X6 A0A097H180 A0A097H143 A0A097H139 A0A097H148 A0A097H169	2.5%
Q38M77	2.4%
A0A097H164 M1AMY0	1.8%

AKV-K2	
AC#	**riBAQ**
Q3S493	9.2%
Q41484 Q2XPX8 Q2XPY0	5.5%
Q3S488 Q3S482 Q3S479	4.1%
Q38M77	1.7%
Q41432 Q3S486 O24372	1.6%
P58515 P24743	1.5%
Q43648 Q07459	1.4%
Q3S485 Q3S483	1.3%

KMC-Feed	
AC#	**riBAQ**
Q3S488 Q3S482 Q3S479	8.3%
Q2XPY0	3.4%
P58515	2.3%
Q8H9D6 Q00652 A0A097H1A0 Q8S382	2.2%
H9B8I9 P17979 P16348 P58520 P58518 H9B8J0 M1AKE5 K7XKR4 A0A097H118 M1AKE3 P58521 C5MRU4 Q43646	1.4%
O24385 Q8RX93 A0A097H166 O24386 Q8RX92	1.0%
A0A097H158 M1AMY0	1.0%

KMC-Food	
AC#	**riBAQ**
Q3S488 Q3S479 Q3S482	12.3%
Q8H9D6 Q00652 A0A097H1A0 Q8S382	7.8%
Q41484 Q2XPX8	6.7%
A0A097H164 M1AMY0	5.3%
A0A097H157	4.6%
P58515 P24743	3.3%
A0A097H120	2.6%
Q38M77	2.2%
O24383 M1KPH5 Q6PMR6 O24384 A0A097H155	1.8%
O24385 Q8RX93 A0A097H166 O24386 Q8RX92	1.6%
Q3S493	1.1%

AKV-PFJ	
AC#	**riBAQ**
Q3S488 Q3S479 Q3S482	8.9%
A0A097H164	7.5%
Q2XPY0	5.8%
H9B8I9 P17979	2.7%
Q8H9D6 Q00652 A0A097H1A0 Q8S382	2.4%
P58515	2.3%
O24383 M1KPH5 Q6PMR6 O24384 A0A097H155	2.1%
A0A097H157	2.0%
A0A097H120	1.4%

Table 6S. Identified and quantified proteins in other classes (non-patatin and non-inhibitor) by LC-MS/MS. Proteins were identified by their UniProt AC# and quantified sample-wise using the iBAQ algorithm in MaxQuant (normalized to riBAQ). [1]Probable heat shock-related protein. [2]Cold-stress inducible protein. [3]Probable metallocarboxypeptidase inhibitor. [4]Histone. [5]Fruit-ripening protein. [6]Probable Probable RNA-binding proteins. [7]Eukaryotic translation initiation factor 5A. [8]Probable Kunitz-type inhibitor (KTI-C type). [9]Probable Pathogenesis-related protein. [10]Probable Kunitz-type inhibitor (KTI-C type). [11]Linoleate 9S-lipoxygenase 1. [12]Probable linoleate 9S-lipoxygenase 3. [13]Probable Kunitz-type inhibitor (KTI-C type).

AKV-Feed		**AKV-K2**		**KMC-Feed**		**KMC-Food**		**AKV-PFJ**	
AC#	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**	**AC#**	**riBAQ**
M1DZV0	1.3%	M1DJ69 M0ZT86 M1DH04 M1AU55 M1AU51 M1AU54 M1AU53 M1AU49 M1AU50	4.3%[1]	M1AMY4 M1AMY7	3.1%[8]	M1AMY4 M1AMY7	1.8%[10]	Q43189 Q9SAP1 Q9SAP0	2.3%[12]
		O04232 M1AM40 Q6ST32 K7X8F1	4.0%[2]	M0ZMA9 M0ZMB0 M0ZMA8 A0A1L1Z6W0	1.0%[9]	P37831	1.1%[11]	M1AMY4 M1AMY7	1.5%[13]
		M1A6J5 M1A6J6 M0ZJ50 O24639	2.9%[3]						
		M1BEW2 M1CM34 M1CM33 M1AG69 M1AG47 Q2XPW1 M1CUG9 M1C576 M1CT06 M1CUH0	2.3%[4]						
		Q2XTC9 M1A8I6 M1A8I5	2.0%[5]						
		M0ZJ84 Q2VCI6 Q38M49 M1D6C4 M1D7Q3 M1D7M9	1.1%[6]						
		Q38HT3	1.0%[7]						

P56337 Q38M78 P56335 P56333 K7VPG0	

Table 7S: Experimental sequence coverage of the selected nine peptides from bioinformatic prediction and *in vitro* functional validation within quantified protein isoforms (patatins, KTI-A, and KTI-B) with 100% identity. Relative abundance (in % riBAQ within each sample) is given for all protein with 100% identity and >90% identity with the respective predicted peptide.

Sample	Experimental coverage	Protein	**Patatin**				**KTI-A**	**KTI-B**			
		Range	**γ-1**	**γ-36**	**α-10**	**α-12**	**β-27**	**β-22**	**γ-38**	**γ-40**	**γ-49**
AKV-Feed	None	0%	0.0%	3.8%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Low	< 25%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Partial	25-75%	0.0%	0.0%	25.3%	0.0%	0.0%	0.0%	5.1%	0.0%	0.0%
	High	>75%	20.2%	0.0%	0.9%	26.2%	0.0%	5.1%	0.0%	5.1%	0.0%
	Full	100%	0.0%	46.5%	0.0%	0.0%	8.8%	0.0%	0.0%	0.0%	0.0%
AKV-K2	None	0%	0.0%	24.5%	0.0%	0.0%	0.0%	5.5%	0.0%	5.5%	0.0%
	Low	< 25%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Partial	25-75%	0.0%	0.0%	14.5%	14.5%	0.0%	0.0%	5.5%	0.0%	0.0%
	High	>75%	9.5%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Full	100%	0.0%	0.0%	0.0%	0.0%	4.1%	0.0%	0.0%	0.0%	0.0%
KMC-Feed	None	0%	0.0%	3.8%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Low	< 25%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Partial	25-75%	0.0%	0.0%	37.1%	0.0%	0.0%	0.0%	3.4%	3.4%	0.0%
	High	>75%	1.1%	0.0%	0.0%	37.1%	0.0%	3.4%	0.0%	0.0%	0.0%
	Full	100%	42.5%	49.9%	0.0%	0.0%	8.3%	0.0%	0.0%	0.0%	0.0%
KMC-Food	None	0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Low	< 25%	0.0%	0.0%	0.0%	0.0%	0.2%	0.1%	0.0%	0.1%	0.0%
	Partial	25-75%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.1%	0.0%	0.1%
	High	>75%	0.0%	0.0%	13.8%	0.0%	0.0%	0.1%	6.7%	0.1%	0.0%
	Full	100%	11.1%	26.0%	0.0%	13.8%	12.3%	6.7%	0.0%	6.7%	0.0%
AKV-PFJ	None	0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%
	Low	< 25%	0.0%	0.0%	0.0%	0.0%	0.0%	0.4%	0.0%	0.4%	0.0%
	Partial	25-75%	0.0%	0.0%	16.3%	16.3%	0.0%	0.0%	0.0%	0.0%	0.0%
	High	>75%	0.7%	0.9%	0.0%	0.0%	0.0%	0.0%	6.2%	0.0%	0.2%
	Full	100%	16.3%	19.6%	0.0%	0.0%	8.9%	5.8%	0.0%	5.8%	0.0%

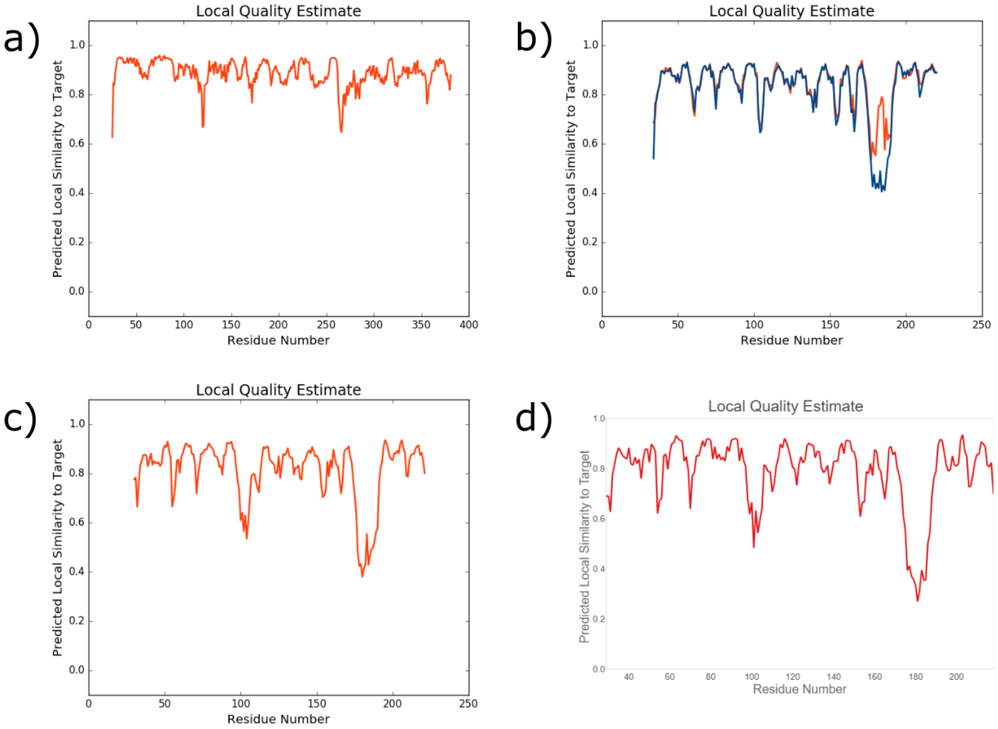

Fig. 1S: Local quality estimates of the template protein models from SwissModel. A) Patatin-B2 (P15477), b) KTI-A (Q3S488), c) KTI-B (Q3S474), and d) KTI-B (Q3S477). QMEAN > 0.75 indicates a good fit between model and template. For KTI-A, QMEAN is shown for both chains in a homo-dimer, where the best fit (chain A, orange) was used for localization of β-27.

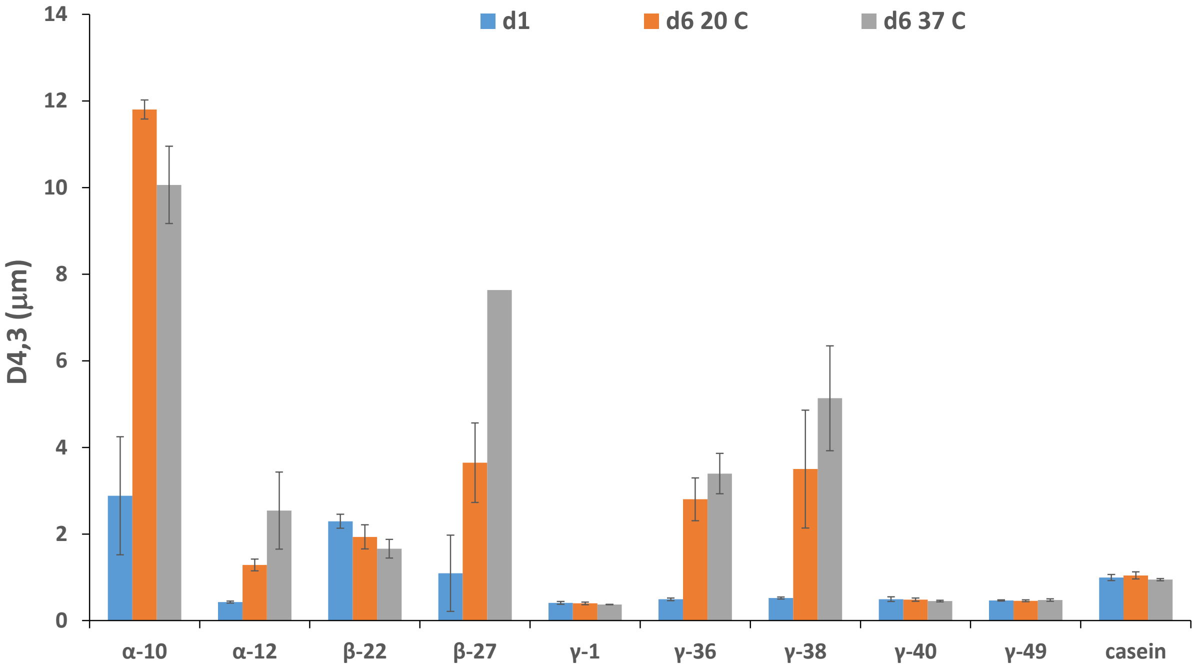

Fig. 2S. Droplet size distribution of emulsions stabilized with selected emulsifier peptides at day 1 and after 6 days storage at 20 °C or 37 °C.