

Quantification of lactoferrin and lactoperoxidase in different types of whey

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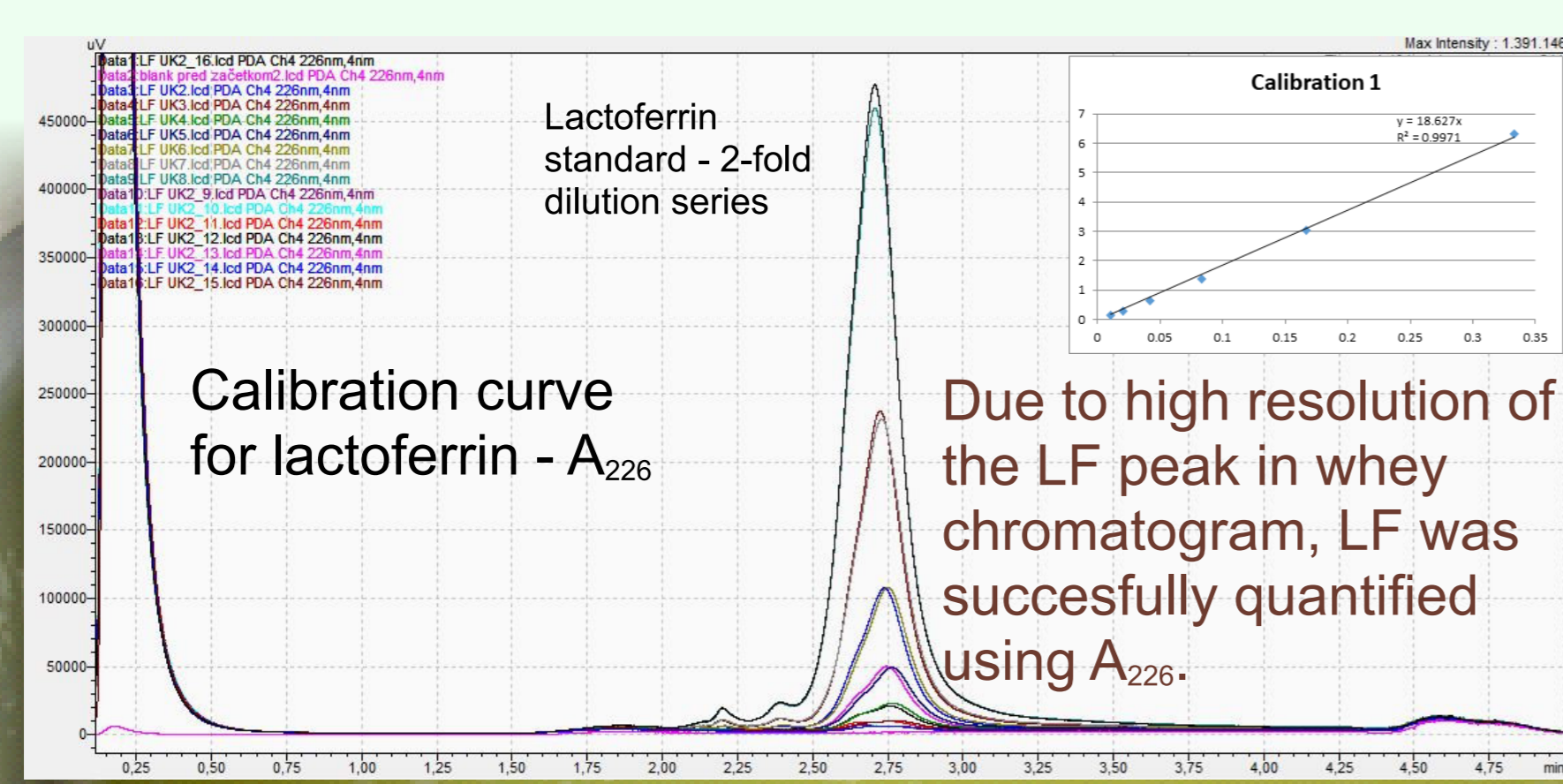
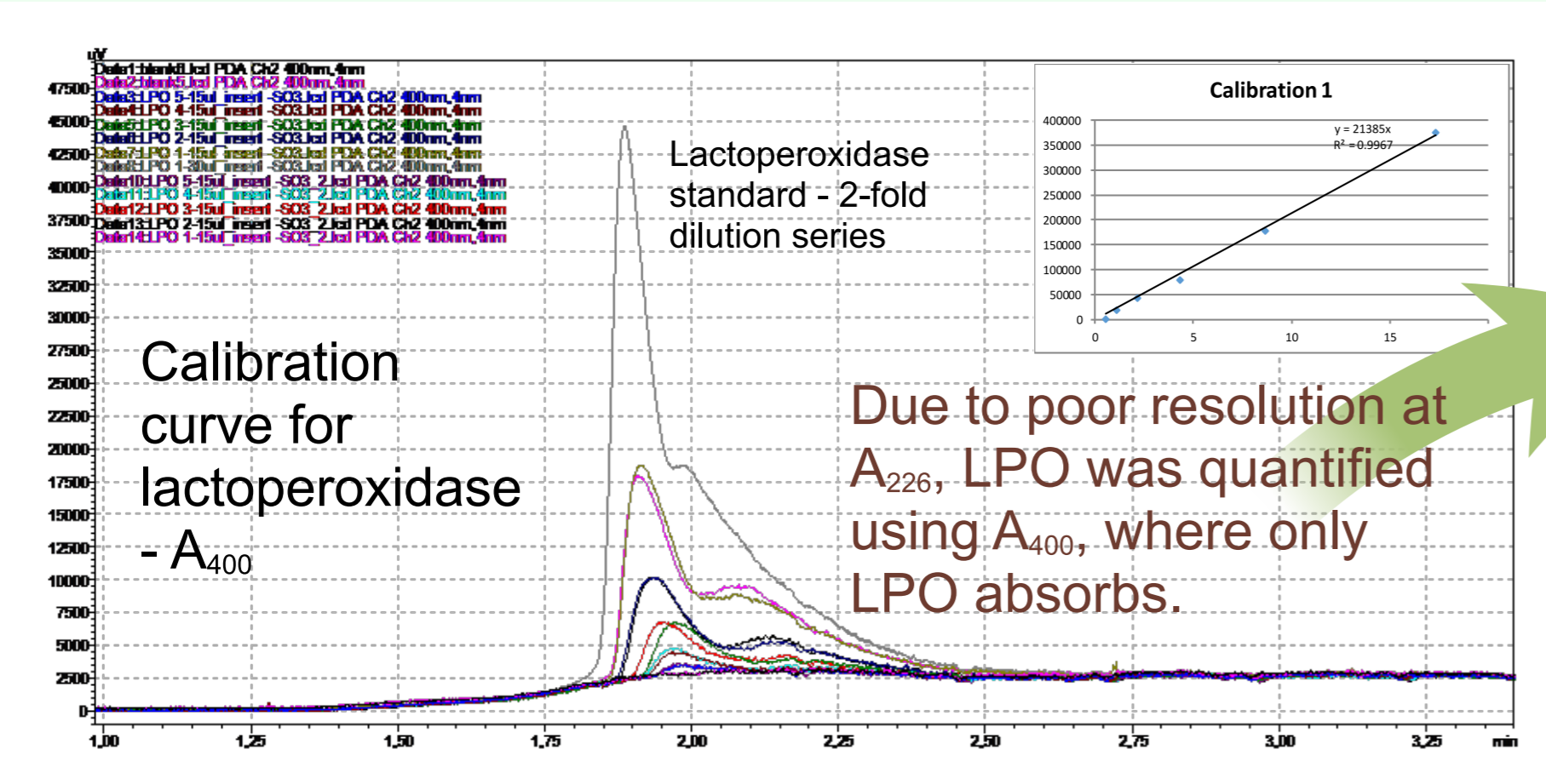
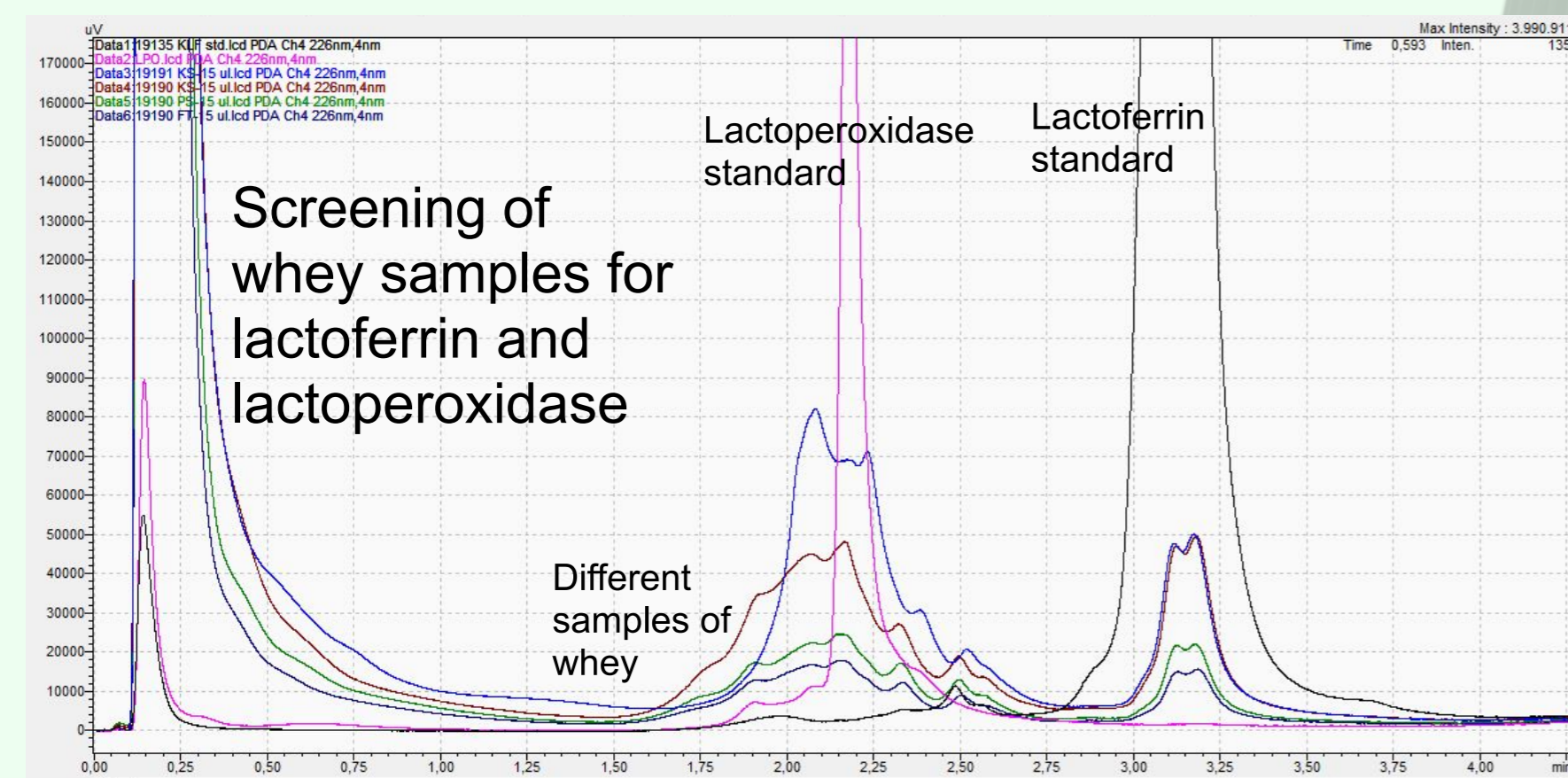
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Introduction

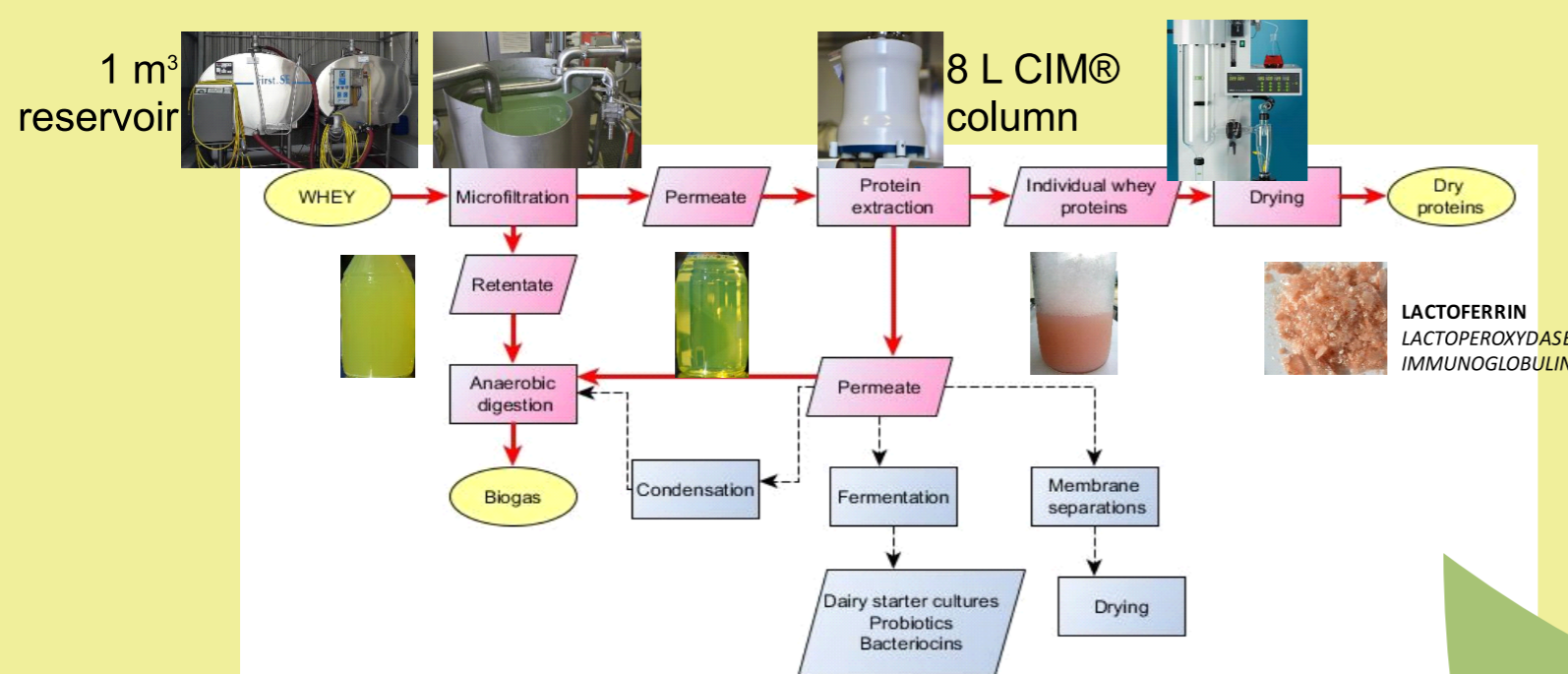
Whey, the liquid by-product in the production of dairy products, represents an abundant potential source of several components of high economic value, e.g. lactose, lactic acid and bioactive proteins such as lactoferrin and lactoperoxidase. Despite the substantial accumulation of knowledge on the usefulness of these components spanning back decades, whey is often still routinely treated as waste and thereby also represents a significant environmental burden.

Methods and results

Cation exchange HPLC



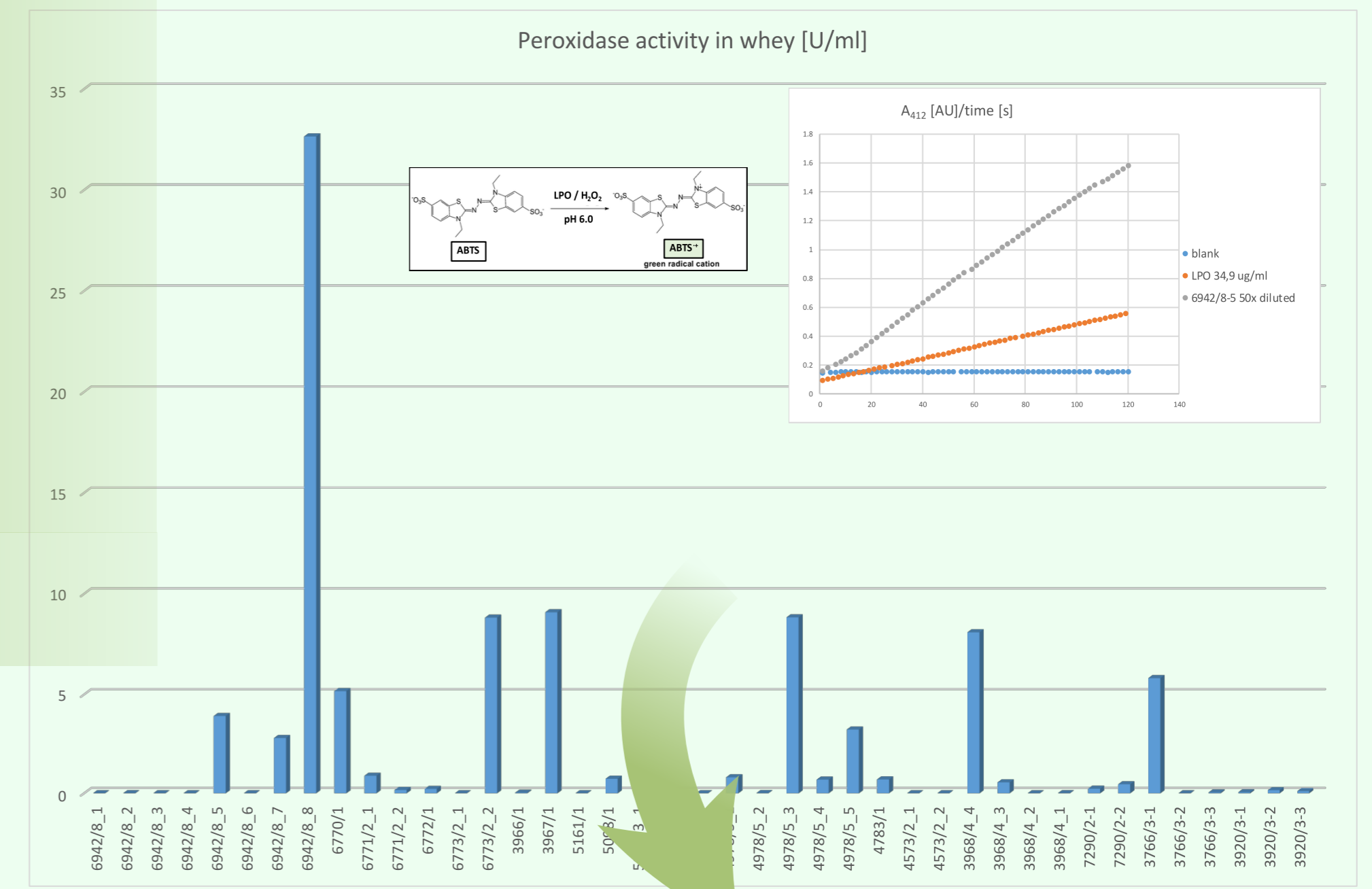
Pilot plant for extraction of proteins at Arhel Ltd.



Acknowledgements

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Peroxidase activity: whey + H₂O₂ + ABTS: A₄₁₂



Sorted by lactoferrin content

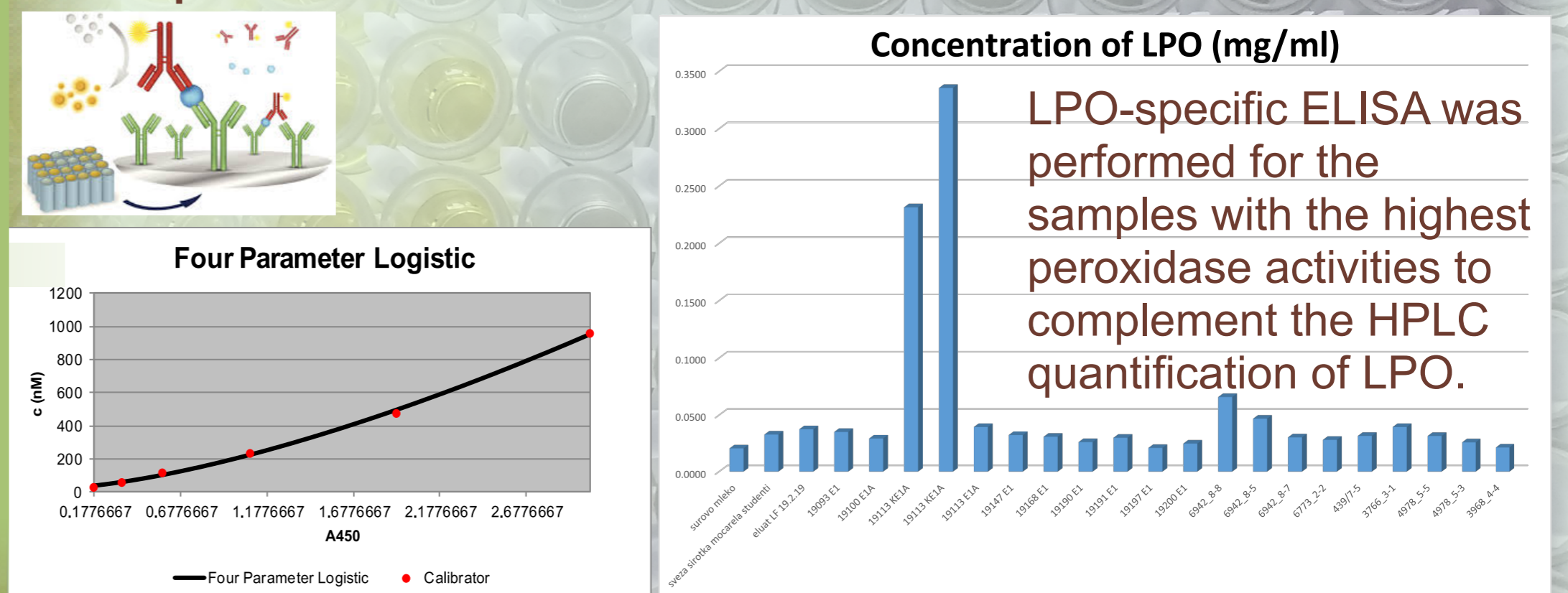
Concentration of LF (mg/ml)	Peroxidase activity (U/ml)	Lab. designation, color coded by type of sample
N/A	0.000	5161/1
N/A	32.651	6942/8-8
0.265	0.325	439/7-2
0.177	0.001	7080/3-3
0.171	5.174	6770/1
0.167	0.202	439/7-1
0.163	0.487	6361/2-1
0.161	0.330	7276/1
0.155	0.169	6771/2-2
0.155	0.001	7320/2-1
0.155	0.000	7080/3-1
0.147	0.099	180/2-1
0.145	0.304	448/1
0.140	0.124	6111/7-2
0.138	0.001	5484/3-1
0.137	0.001	1250/1
0.136	0.095	130/1
0.135	0.071	6111/7-1
0.135	0.048	1302/2-1
0.134	0.044	3766/3-3
0.134	0.000	5484/3-2
0.132	0.013	871/2-2
0.132	0.002	1737/3-1
0.132	0.001	488/3-1
0.131	0.111	3920/3-3
0.130	0.002	488/3-2
0.126	0.002	1737/3-2
0.112	0.456	7290/2-2
0.112	0.029	3966/1
0.103	0.734	5098/1

Sorted by peroxidase activity

Concentration of LF (mg/ml)	Peroxidase activity (U/ml)	Concentration of LPO - HPLC (mg/ml)	Concentration of LPO - ELISA (mg/ml)	Specific activity - HPLC (U/mg)	Specific activity - ELISA (U/mg)	Lab. designation, color coded by type of sample
N/A	32.651	0.386	0.066	84.56	495.21	6942/8-8
0.094	10.480	0.137	0.032	76.39	332.05	439/7-5
0.081	9.086	0.174	N/A	52.27	N/A	3967/1
0.081	8.834	0.170	0.026	51.95	341.53	4978/5-3
0.070	8.820	0.094	0.028	93.61	314.47	6773/2-2
0.030	8.094	0.062	0.021	129.68	380.21	3968/4-4
0.029	5.825	0.090	0.039	64.91	147.80	3766/3-1
0.171	5.174	0.056	N/A	91.83	N/A	6770/1
0.070	3.911	0.081	0.047	48.03	83.61	6942/8-5
0.000	3.226	0.089	0.031	36.05	102.45	4978/5-5
0.032	2.798	0.089	0.030	31.28	92.48	6942/8-7
0.076	0.886	0.011	N/A	80.99	N/A	6771/2-1
0.036	0.808	0.035	N/A	22.85	N/A	4978/5-1
0.103	0.734	0.020	N/A	36.74	N/A	5098/1
0.058	0.725	0.015	N/A	49.32	N/A	6361/2-2
0.019	0.700	0.031	N/A	22.56	N/A	4783/1
0.040	0.693	0.046	N/A	14.90	N/A	4978/5-4
0.000	0.553	0.054	N/A	10.32	N/A	3968/4-3
0.000	0.497	0.084	N/A	5.94	N/A	Zer Cottage
0.163	0.487	0.039	N/A	12.60	N/A	6361/2-1
0.059	0.478	0.029	N/A	16.63	N/A	439/7-6
0.112	0.456	0.018	N/A	25.89	N/A	7290/2-2
0.161	0.330	0.028	N/A	11.93	N/A	7276/1
0.265	0.325	0.069	N/A	4.70	N/A	439/7-2
0.145	0.304	0.023	N/A	13.04	N/A	448/1
0.038	0.247	0.027	N/A	9.21	N/A	439/7-7
0.024	0.232	0.022	N/A	10.39	N/A	1302/2-2
0.099	0.230	0.018	N/A	12.82	N/A	7290/2-1
0.075	0.226	0.022	N/A	10.41	N/A	871/2-1
0.096	0.219	0.174	N/A	1.26	N/A	6772/1

Up to 0.17 g/L of lactoferrin was detected in acid whey derived from fresh curd cheese production, and up to 0.10 g/L in sweet whey obtained in cheese production. In 42 % of samples, lactoferrin concentration was below the detection limit. The enzymatic activity of lactoperoxidase reached up to 10 U/mL in sweet cheese whey, while in 32 % of samples lactoperoxidase was not detected. The concentration of peroxidase reached up to 0.174 g/L (HPLC) and the calculated specific activity up to 130 U/mg in non-concentrated samples of whey.

Lactoperoxidase ELISA



Discussion

The samples of acid whey from fresh curd cheese proved to be the most promising in the exploitation of lactoferrin, whereas sweet whey obtained in cheese production seems to have the highest potential for the isolation of enzymatically active lactoperoxidase. Our results will be used as means of zeroing in on the dairy plants and technologies that are "good producers" of these proteins. The further goal is to evaluate the possibility of advanced processing of whey, which would offer dairies an additional income and reduce the environmental impact of whey-derived waste. The separation of lactoferrin and lactoperoxidase is already ongoing also in a demonstration plant (10000 L/day) based on CIM® monolithic ion-exchange chromatography columns in the frame of LIFE for Acid Whey (LIFE16 ENV/SI/000335) project.