

ANNEX

LIPID ANALYSIS

Table 44. Total content of extracted lipids – EpiDerm model.

	Batch 1	Batch 2	Batch 3	Mean	SD
Surface of the model (cm ²)	0.63	0.63	0.63		
Weight of the dry sample (mg)	1.9	2.2	2.3	2.13	0.21
Total content of extracted lipids (mg)	0.242	0.258	0.288	0.263	0.023
Total lipid content expressed as mg/cm ²	0.384	0.410	0.456	0.417	0.037
Lipid content expressed as % of sample dry weight	12.72	11.74	12.50	12.321	0.514

Table 45. Percentile content of individual lipid classes extracted from EpiDerm model.

TLC detected lipids						Calculation per dry matter					
	B1	B2	B3	mean	SD	B1	B2	B3	mean	SD	
	%	%	%	%		%	%	%	%		
CE	7.4	3.4	8.4	6.4	2.6	CE	0.74	0.19	0.61	0.52	0.29
TG	7.1	4.3	2.9	4.8	2.2	TG	0.72	0.24	0.21	0.39	0.28
CHOL	34.9	40.4	32.1	35.8	4.2	CHOL	3.52	2.30	2.36	2.73	0.69
FFA	5.9	5.7	10.2	7.3	2.6	FFA	0.60	0.32	0.75	0.56	0.22
Ceramides together	15.3	23.3	17.0	18.5	4.2	Ceramides together	1.35	1.46	1.29	1.37	0.09
Cerebrozides together	28.4	21.9	28.0	26.1	3.7	Cerebrozides together	2.86	1.24	2.06	2.05	0.81
CHOL SO ₄	0.9	1.0	1.4	1.1	0.2	CHOL SO ₄	0.09	0.06	0.10	0.08	0.02
Sum	100.0	100.0	100.0	100.0		Sum	9.87	5.82	7.39	7.69	2.04

B – batch, SD – standard deviation.

CE - cholesterol esters, TG - triglycerides, CHOL- cholesterol, FFA - free fatty acids, CHOL SO₄ - cholesterol sulphate.

Table 46. Total content of extracted lipids – EPISKIN model.

	batch 1	batch 2	batch 3	mean	SD
Surface (cm ²)	1.1	1.1	1.1		
Weight of the dry sample (mg)	6.4	6.8	5.9	6.4	0.45
Total content of extracted lipids (mg)	0.700	0.688	0.604	0.664	0.052
Total lipid content expressed as mg/cm ²	0.636	0.625	0.549	0.604	0.047
Lipid content expressed as % of sample dry weight	10.94	10.11	10.24	10.429	0.445

Table 47. Percentile content of individual lipid classes extracted from EPISKIN model.

TLC detected lipids						Calculation per dry matter					
	B1	B2	B3	mean	SD	B1	B2	B3	mean	SD	
	%	%	%	%		%	%	%	%		
CE	9.8	5.9	7.5	7.7	2.0	CE	0.80	0.46	0.74	0.67	0.18
TG	3.0	4.4	2.7	3.4	1.0	TG	0.24	0.34	0.26	0.28	0.05
CHOL	41.1	41.8	49.0	44.0	4.4	CHOL	3.35	3.23	4.85	3.81	0.90
FFA	9.2	9.4	10.4	9.7	0.7	FFA	0.75	0.73	1.03	0.84	0.17
Ceramides together	25.1	23.6	21.2	23.3	2.0	Ceramides together	2.20	2.02	2.44	2.22	0.21
Cerebrozides together	11.1	14.1	8.7	11.3	2.7	Cerebrozides together	0.90	1.09	0.86	0.95	0.12
CHOL SO ₄	0.7	0.7	0.5	0.6	0.1	CHOL SO ₄	0.06	0.05	0.05	0.05	0.01
Summ	100.0	100.0	100.0	100.0		Summ	8.31	7.92	10.24	8.82	1.24

B – batch, SD – standard deviation.

CE - cholesterol esters, TG - triglycerides, CHOL- cholesterol, FFA - free fatty acids, CHOL SO₄ - cholesterol sulphate.

Table 48. Total content of extracted lipids – SkinEthic model.

	batch 1	batch 2	batch 3	mean	SD
Surface (cm ²)	0.5	0.5	0.5		
Weight of the dry sample (mg)	2.5	1.8	2.5	2.27	0.4
Total content of extracted lipids (mg)	0.319	0.279	0.283	0.294	0.022
Total lipid content expressed as mg/cm ²	0.637	0.558	0.567	0.587	0.043
Lipid content expressed as % of sample dry weight	12.75	15.51	11.332	13.197	2.125

Table 49. Percentile content of individual lipid classes extracted from SkinEthic model.

TLC detected lipids	B1	B2	B3	mean	SD	Calculation per dry matter					
						B1	B2	B3	mean	SD	
	%	%	%	%		%	%	%	%		
CE	7.4	9.4	7.3	8.0	1.2	CE	0.63	0.82	0.50	0.65	0.16
TG	1.9	2.8	2.5	2.4	0.4	TG	0.16	0.24	0.17	0.19	0.04
CHOL	42.9	30.8	49.0	40.9	9.3	CHOL	3.64	2.67	3.35	3.22	0.50
FFA	14.2	10.2	11.7	12.1	2.0	FFA	1.21	0.89	0.80	0.97	0.22
Ceramides together	26.6	38.6	22.1	29.1	8.5	Ceramides together	3.28	3.08	2.08	2.81	0.64
Cerebrozides together	6.2	7.3	7.0	6.8	0.6	Cerebrozides together	0.53	0.63	0.48	0.55	0.08
CHOL SO ₄	0.8	0.9	0.3	0.7	0.3	CHOL SO ₄	0.06	0.08	0.02	0.06	0.03
Summ	100.0	100.0	100.0	100.0		Summ	9.52	8.40	7.40	8.44	1.06

B – batch, SD – standard deviation,

CE - cholesterol esters, TG - triglycerides, CHOL- cholesterol, FFA - free fatty acids, CHOL SO₄ - cholesterol sulphate,

TEER RAW DATA

Table 50. Results obtained in the TEER assay and time-course assay with Triton 1% - EpiDerm model.

Exposure	Raw data (k Ω)				TEER (k Ω .cm ²)				TEER decrease %	Tissue viability %	Tissue difference %
	Before exposure		After exposure		Before exposure		After exposure				
	mean	sd	mean	sd	mean	sd	mean	sd			
Run 1											
NC (4h)	0.66	0.01	0.70	0.04	0.34	0.01	0.36	0.02	106.48	100.0	0.1
2 h	0.75	0.05	0.33	0.00	0.39	0.03	0.13	0.00	33.60	90.6	1.1
4 h	0.97	0.01	0.24	0.04	0.54	0.01	0.07	0.02	13.53	76.8	6.4
6 h	0.93	0.44	0.12	0.00	0.51	0.28	0.00	0.00	0.00	21.9	7.9
8 h	1.04	0.24	0.12	0.00	0.58	0.15	0.00	0.00	0.00	12.1	0.9
10 h	1.19	0.08	0.12	0.00	0.67	0.05	0.00	0.00	0.00	10.2	0.9
Run 2											
NC (4h)	0.72	0.04	0.90	0.01	0.38	0.03	0.49	0.00	129.17	100.0	1.5
2 h	0.78	0.01	0.33	0.03	0.42	0.01	0.13	0.02	31.82	93.8	1.1
4 h	0.75	0.25	0.28	0.04	0.39	0.16	0.10	0.02	24.80	76.2	3.4
6 h	0.98	0.01	0.15	0.00	0.54	0.00	0.02	0.00	3.51	33.3	8.8
8 h	0.78	0.25	0.13	0.01	0.42	0.16	0.00	0.00	0.76	14.4	0.9
10 h	1.02	0.14	0.13	0.00	0.57	0.09	0.01	0.00	1.11	12.7	0.5
Run 3											
NC (4h)	0.69	0.07	0.72	0.04	0.36	0.04	0.38	0.03	105.26	100.0	0.8
2 h	0.98	0.01	0.34	0.00	0.54	0.01	0.14	0.00	25.58	93.6	0.1
4 h	1.35	0.08	0.28	0.04	0.77	0.05	0.10	0.02	12.60	88.9	3.3
6 h	0.98	0.25	0.18	0.04	0.54	0.16	0.03	0.02	6.40	80.9	5.2
8 h	0.96	0.27	0.12	0.00	0.53	0.17	0.00	0.00	0.00	23.2	4.5
10 h	1.10	0.08	0.12	0.00	0.61	0.05	0.00	0.00	0.00	16.0	1.8

Table 51. Results obtained in the TEER assay and time-course assay with Triton 1% - EPISKIN model.

Exposure	Raw data (k Ω)				TEER (k Ω .cm ²)				TEER decrease %	Tissue viability %	Tissue difference %
	Before exposure		After exposure		Before exposure		After exposure				
	mean	sd	mean	sd	mean	sd	mean	sd			
Run 1											
NC (4h)	16.60	4.81	14.80	5.52	18.26	5.29	16.28	6.07	89.16	100.0	8.1
2 h	15.00	3.54	10.28	1.60	16.50	3.89	11.31	1.76	68.53	118.3	7.9
4 h	5.26	3.24	5.19	0.28	5.79	3.56	5.70	0.30	98.57	96.3	0.4
6 h	6.54	4.64	3.46	0.75	7.19	5.10	3.81	0.82	52.91	109.4	0.4
8 h	17.72	0.75	2.15	0.31	19.49	0.82	2.37	0.34	12.13	98.6	2.7
10 h	13.92	6.53	1.80	0.17	15.31	7.18	1.98	0.19	12.94	28.8	0.8
Run 2											
NC (4h)	11.24	1.76	10.82	0.52	12.36	1.94	11.90	0.57	96.26	100.0	1.2
2 h	8.25	0.64	4.95	2.47	9.08	0.70	5.45	2.72	60.00	103.5	2.7
4 h	16.65	4.74	13.30	7.07	18.32	5.21	14.63	7.78	79.88	94.0	6.0
6 h	16.73	2.81	8.71	0.16	18.40	3.10	9.58	0.17	52.06	78.2	0.1
8 h	11.11	4.94	4.32	1.82	12.22	5.44	4.75	2.00	38.86	57.6	0.4
10 h	14.66	5.58	1.65	0.68	16.12	6.14	1.82	0.75	11.26	15.8	1.5
Run 3											
NC (4h)	4.65	0.32	2.81	0.74	5.11	0.35	3.09	0.82	60.39	100.0	0.8
2 h	6.28	3.34	3.83	1.71	6.90	3.68	4.21	1.88	61.04	93.6	0.1
4 h	6.55	3.18	4.36	1.49	7.21	3.50	4.79	1.64	66.49	88.9	3.3
6 h	3.27	1.94	1.74	0.57	3.60	2.13	1.91	0.62	53.21	80.9	5.2
8 h	4.71	0.30	1.52	0.11	5.18	0.33	1.67	0.12	32.27	23.2	4.5
10 h	7.35	2.90	1.34	0.23	8.09	3.19	1.47	0.26	18.16	16.0	1.8

Table 52. Results obtained in the TEER assay and time-course assay with Triton 1% - SkinEthic model.

Exposure	Raw data (k Ω)				TEER (k Ω .cm ²)				TEER decrease %	Tissue viability %	Tissue difference %
	Before exposure		After exposure		Before exposure		After exposure				
	mean	sd	mean	sd	mean	sd	mean	sd			
Run 1											
NC (4h)	20.00	0.00	20.00	0.00	9.95	0.00	9.95	0.00	100.00	100.0	0.1
2 h	20.00	0.00	1.40	0.47	9.95	0.00	0.64	0.24	6.46	84.3	1.9
4 h	20.00	0.00	0.76	0.01	9.95	0.00	0.33	0.01	3.27	64.4	4.3
6 h	20.00	0.00	0.74	0.26	9.95	0.00	0.31	0.13	3.14	38.5	10.2
8 h	18.16	2.60	0.56	0.07	9.03	1.30	0.23	0.04	2.49	9.7	0.1
10 h	20.00	0.00	0.43	0.11	9.95	0.00	0.16	0.06	1.61	6.8	0.6
Run 2											
NC (4h)	20.00	0.00	20.00	0.00	9.95	0.00	9.95	0.00	100.00	100.0	0.6
2 h	20.00	0.00	1.42	0.14	9.95	0.00	0.88	0.12	6.59	73.0	2.5
4 h	20.00	0.00	0.59	0.30	6.59	4.51	0.42	0.14	2.39	40.1	3.4
6 h	9.65	9.04	0.33	0.04	7.12	3.84	0.16	0.04	2.25	11.2	0.2
8 h	18.16	2.60	0.25	0.04	9.95	0.00	0.14	0.02	0.78	5.6	0.2
10 h	20.00	0.00	0.22	0.04	9.95	0.00	0.09	0.01	0.55	5.3	0.3
Run 3											
NC (4h)	20.00	0.00	20.00	0.00	10.00	14.14	10.00	14.14	100.00	100.0	1.8
2 h	20.00	0.00	1.87	0.23	10.00	14.14	1.05	1.15	8.82	83.9	0.5
4 h	13.28	9.02	0.95	0.29	11.15	3.01	0.62	0.46	6.34	73.8	1.9
6 h	14.36	7.69	0.43	0.08	11.02	4.72	0.25	0.25	2.21	35.7	3.5
8 h	20.00	0.00	0.40	0.04	10.00	14.14	0.22	0.25	1.43	12.8	0.8
10 h	20.00	0.00	0.29	0.02	10.00	14.14	0.15	0.19	0.88	7.0	0.1