Appendix No. 1 – ML 2340/21

Records documenting analysis of the sample using U-HPLC-HRMS/MS metabolomic profiling (targeted screening) method

Sample description

Lab code	Sample name (provided by the client)	Sample description
ML 2340/21	Olejek konopny 15 % Expiration date: 18.6.2022	Hemp extract in oil

Testing strategy

Metabolomic profiling (targeted screening) strategy was selected for the testing of the sample (laboratory code ML 2340/21). This type of analytical approach is based on UCT internal databases of compounds which may occur in *Cannabis sativa* L. and products thereof; the first UCT database (A) includes molecular spectral information for 244 minor phytocannabinoids, the second UCT database (B) includes molecular spectral information for 151 other (non-cannabinoid) biologically active compounds (e.g. terpenoids, phenols, bibenzyl stilbenes, fatty acids, amides, flavones, lignans, flavonoid glycosides, lignanamide derivatives). The analysis of given sample was performed by ultra-high performance liquid chromatography coupled to high resolution tandem mass spectrometry (ISO 17025 accredited method KM 15, system E: U-HPLC-HRMS/MS (Q-TOF)). Detailed description of analytical procedures and conditions (SOP) are available at Laboratory.

Testing conditions

Sample preparation consisted of sample dissolution in ethanol and following dilution. Subsequently, a reversed-phase chromatographic column was used for the compound's separation; quadrupole/time of flight mass analyzer (Agilent 6560 Q-TOF) with electrospray ionization source was used for their detection. Each sample was injected into U-HPLC – HRMS/MS system in several dilutions to overcome matrix effects including column and/or detector saturation for particular compounds. The data were recorded and evaluated separately for both the positive and negative electrospray ionization mode (ESI+ and ESI-). For the data processing, Agilent MassHunter Profinder and Agilent MassHunter Qualitative Analysis softwares were used.

Test results and interpretation

The overall chemical compositions of the sample detectable under given method conditions are illustrated by total compound chromatograms (TCC) (**Figure 1 - 2**) recorded in ESI+ and ESI- ionization mode. The TCC shows all features (compounds) detected in the sample, non-filtered by the UCT databases.

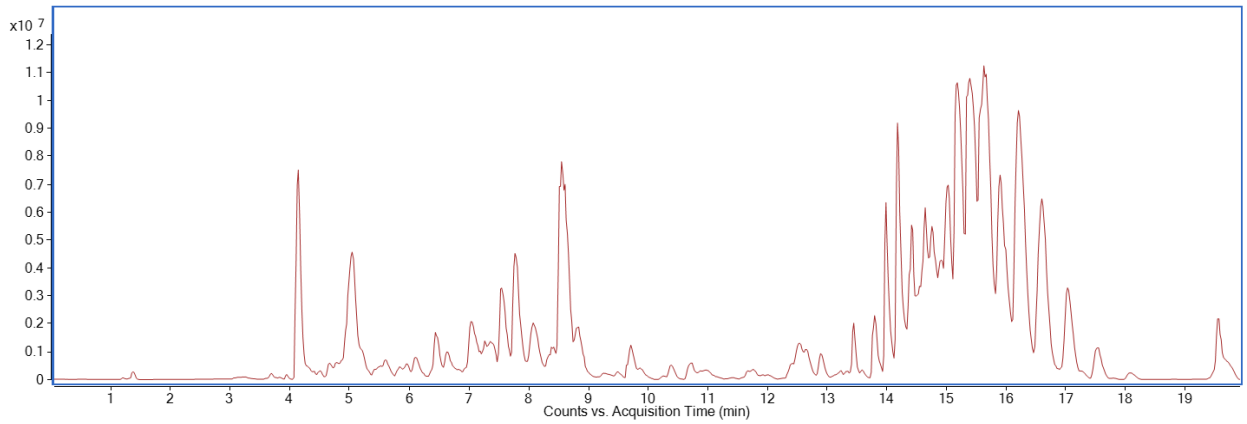

Figure 1: ML 2340/21 - UHPLC-HRMS/MS total compound chromatogram obtained for undiluted sample in ESI+

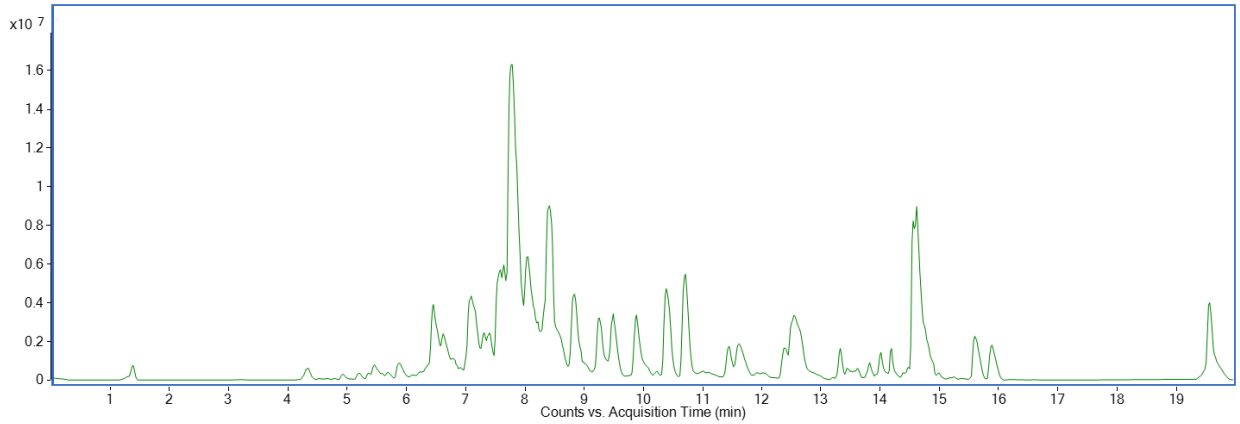

Figure 2: ML 2340/21 - UHPLC-HRMS/MS total compound chromatogram obtained for undiluted sample in ESI-

A) Targeted screening of minor phytocannabinoids

In the tested sample, the following amounts of minor phytocannabinoids (**Table I**) characterized by unique combination of exact mass and retention time RT were detected.

Table I: Number of minor phytocannabinoids detected in sample in ESI+ and ESI-

Sample name	Number of minor phytocannabinoids detected in ESI+	Number of minor phytocannabinoids detected in ESI-
ML 2340/21	4	56

Extracted ion chromatograms (XIC) of the detected compounds in both ionization modes (ESI+, ESI-) are documented on **Figures 3 - 4**. All detected phytocannabinoids (tentative identification) are also

summarized in **Table II** (ESI+) and **Table III** (ESI-). Considering the possible existence of isomeric forms, one detected compound may have several identities.

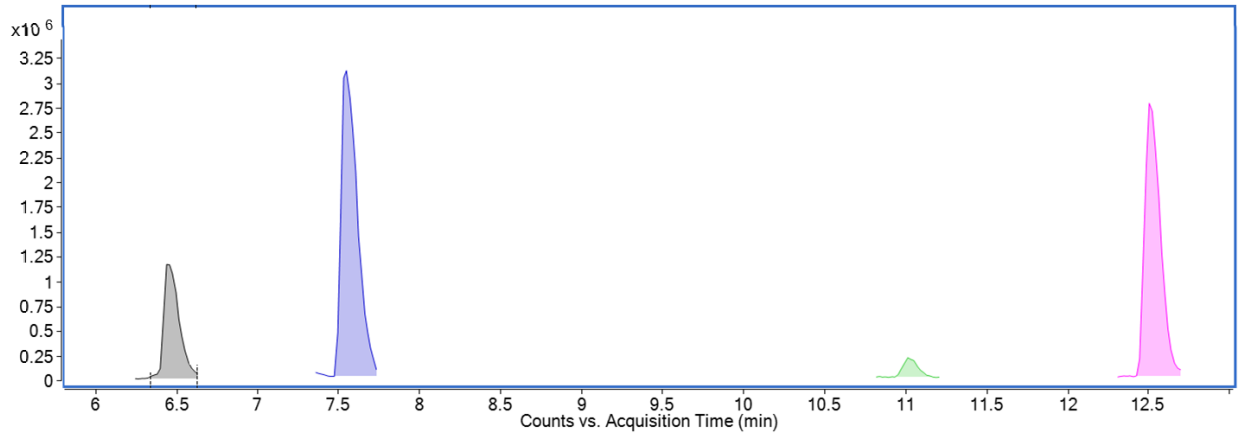

Figure 3: ML 2340/21 - UHPLC-HRMS/MS extracted ion chromatogram of 4 compounds with unique combination of exact mass and RT detected in ESI+ (obtained for undiluted sample)

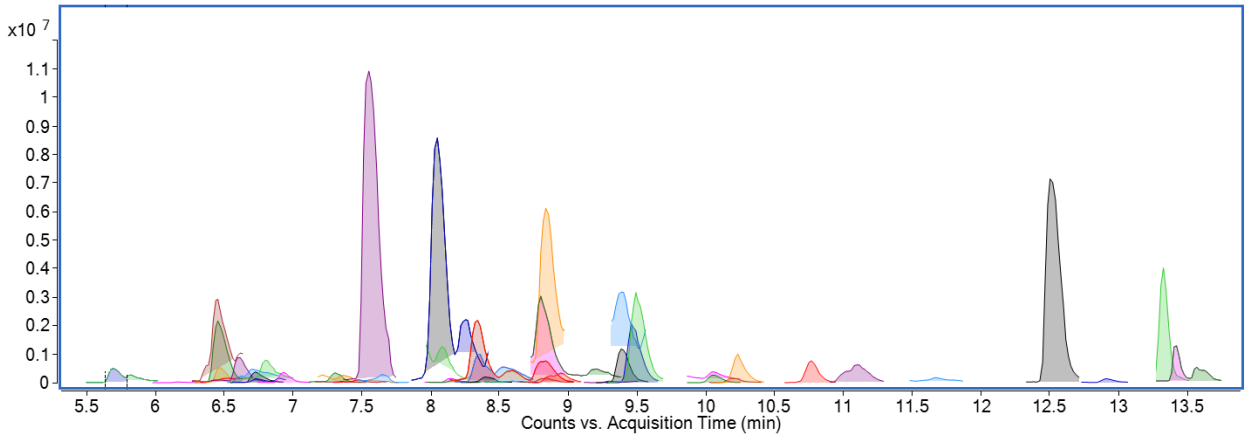

Figure 4: ML 2340/21 - UHPLC-HRMS/MS extracted ion chromatogram of 56 compounds with unique combination of exact mass and RT detected in ESI- (obtained for undiluted sample)

Table II: Overview of compounds detected in ESI+ (corresponding with the Figures 3)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Peak area[e]
1	374.2076	C22 H30 O5	6.44	Cannabielsoic acid A/cannabielsoic acid B	2.8E+06
2	332.2334	C21 H32 O3	7.54	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	2.3E+07
3	314.2229	C21 H30 O2	11.01	(1aS.3aR.8bR.8cR)-Cannabicyclol/cannabicitran/(-)-delta9-cis-(6aS.10aR)-delta9-tetrahydrocannabinol/(âˆ’)-delta7 -trans-(1R. 3R. 6R)-isotetrahydrocannabinol-C5/cannabigeroquinone	1.4E+06
4	640.4485	C43 H60 O4	12.50	Cannabisol	5.6E+06

[a] *Mass error compared the theoretical exact mass < 5 ppm*

[b] *Match of isotopic pattern confirmed*

[c] *Retention time*

[d] *Tentative identification of compounds based on available scientific articles (Hanuš et al. 2016, Mechoulam 2002); possible identities are separated by the symbol "/"*

[e] *Peak area related to the undiluted sample prepared according to stated procedure*

Table III: Overview of compounds detected in ESI- (corresponding with the Figure 4)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Peak area[e]
1	330.1830	C20 H26 O4	5.69	Cannabichromevarinic acid/delta-9-tetrahydrocannabivarinic acid	2.9E+06
2	330.1831	C20 H26 O4	5.82	Cannabichromevarinic acid/delta-9-tetrahydrocannabivarinic acid	7.8E+05
3	350.2452	C21 H34 O4	6.17	Carmagerol	1.8E+05
4	282.1624	C19 H22 O2	6.45	Cannabinodivarin/cannabivarin/demethyldecarboxyamorfrutin A/63b/63c	1.4E+07
5	328.2041	C21 H28 O3	6.45	Cannabichromanone-D/cannabicoumaronone/10-oxo-delta-6a(10a)-tetrahydrocannabinol/8-Oxo-delta9-trans-tetrahydrocannabinol/9.10-Anhydrocannabitriol/anhydrocannabimovone/Cannabidiol Hydroxyquinone	3.9E+06
6	302.1522	C18 H22 O4	6.47	delta-9-Tetrahydrocannabiorcolic acid A/delta-9-tetrahydrocannabiorcolic acid B/cannabiorcichromenic acid/anthopogochromenic acid/cannabiorcicyclolic acid/anthopogocyclolic acid	3.0E+06
7	288.2078	C19 H28 O2	6.51	Cannabigerovarin	1.1E+06
8	376.2251	C22 H32 O5	6.60	CA30/ CA31 rac-6'-Epoxycannabigerolic acid (2'S*. 3'R*)/rac-6'-epoxycannabigerolic acid (2'R*. 3'R*)	5.3E+06
9	332.2349	C21 H32 O3	6.71	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	5.6E+06
10	354.1834	C22 H26 O4	6.73	Cannabinolic acid A	2.5E+06
11	304.2035	C19 H28 O3	6.75	Cannabiglendol C3	7.3E+05
12	348.2299	C21 H32 O4	6.81	Cannabiripsol	3.4E+06
13	258.1624	C17 H22 O2	6.93	Cannabidiorcol/delta-9-trans-tetrahydrocannabiorcol/cannabiorcitran/cannabiorcicyclol/cannabiorcichromene	2.0E+06
14	330.2200	C21 H30 O3	7.27	8'-Hydroxyisocannabichromene /cannabielsoin/abnormal cannabigeroquinol/10alpha-hydroxy trans delta-8-tetrahydrocannabinol/10 beta-hydroxy trans delta-8-tetrahydrocannabinol/8 alpha-hydroxy-delta9-trans-tetrahydrocannabinol/8 beta-hydroxy-delta9-trans-tetrahydrocannabinol/tetrahydrocannabinol epoxide/hydroxy delta9.11-hexahydrocannabinol	1.0E+07
15	362.1883	C24 H26 O3	7.31	Machaeridiol C/machaeriol B	2.0E+06
16	304.2034	C19 H28 O3	7.35	Cannabiglendol C3	2.9E+05
17	348.2297	C21 H32 O4	7.37	Cannabiripsol	1.4E+06

Table III - continuation: Overview of compounds detected in ESI- (corresponding with the Figure 4)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Peak area[e]
18	258.1624	C17 H22 O2	7.48	Cannabidiorcol/delta-9-trans-tetrahydrocannabiorcol/cannabiorcitran/cannabiorcicyclol/cannabiorcichromene	4.4E+05
19	332.2356	C21 H32 O3	7.55	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	8.3E+07
20	348.2287	C21 H32 O4	7.58	Cannabiripsol	5.9E+05
21	288.2062	C19 H28 O2	7.65	Cannabigerovarin	1.5E+06
22	330.2200	C21 H30 O3	8.05	8'-Hydroxyisocannabichromene /cannabielsoin/abnormal cannabigeroquinol/10alpha-hydroxy trans delta-8-tetrahydrocannabinol/10 beta-hydroxy trans delta-8-tetrahydrocannabinol/8 alpha-hydroxy-delta9-trans-tetrahydrocannabinol/8 beta-hydroxy-delta9-trans-tetrahydrocannabinol/tetrahydrocannabinol epoxide/hydroxy delta9.11-hexahydrocannabinol	5.8E+07
23	300.2081	C20 H28 O2	8.09	delta-9-Tetrahydrocannabinol-C4/norCannabidiol	8.8E+06
24	282.1622	C19 H22 O2	8.15	Cannabinodivarin/cannabivarin/demethyldecarboxyamorfrutin A/63b/63c	8.6E+05
25	258.1620	C17 H22 O2	8.17	Cannabidiorcol/delta-9-trans-tetrahydrocannabiorcol/cannabiorcitran/cannabiorcicyclol/cannabiorcichromene	5.8E+05
26	330.2198	C21 H30 O3	8.27	8'-Hydroxyisocannabichromene /cannabielsoin/abnormal cannabigeroquinol/10alpha-hydroxy trans delta-8-tetrahydrocannabinol/10 beta-hydroxy trans delta-8-tetrahydrocannabinol/8 alpha-hydroxy-delta9-trans-tetrahydrocannabinol/8 beta-hydroxy-delta9-trans-tetrahydrocannabinol/tetrahydrocannabinol epoxide/hydroxy delta9.11-hexahydrocannabinol	1.0E+07
27	332.2354	C21 H32 O3	8.34	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	1.5E+07
28	346.2142	C21 H30 O4	8.37	Trans-10-ethoxy-9-hydroxy-delta6a(10a)-tetrahydrocannabivarin-C3/cannabimovone/(+)-(9S.10S)-trans-cannabitriol/(-)-(9R.10R)-trans-cannabitriol/(9S.10R)-cis-cannabitriol/(9R.10S)-cis-cannabitriol/ethoxy-cannabitriolvarin/isocannabitriol (8.9-dihydroxy-delta-6a(10a)-tetrahydrocannabinol)	1.6E+07
29	288.2078	C19 H28 O2	8.40	Cannabigerovarin	1.4E+06

Table III - continuation: Overview of compounds detected in ESI- (corresponding with the Figure 4)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Peak area[e]
30	346.2145	C21 H30 O4	8.55	Trans-10-ethoxy-9-hydroxy-delta6a(10a)-tetrahydrocannabivarin-C3/cannabimovone/(+)-(9S.10S)-trans-cannabitriol/(-)-(9R.10R)-trans-cannabitriol/(9S.10R)-cis-cannabitriol/(9R.10S)-cis-cannabitriol/ethoxy-cannabitriolvarin/isocannabitriol (8.9-dihydroxy-delta-6a(10a)-tetrahydrocannabinol)	2.0E+07
31	258.1619	C17 H22 O2	8.59	Cannabidiorcol/delta-9-trans-tetrahydrocannabiorcol/cannabiorcitran/cannabiorcicyclol/cannabiorcichromene	4.6E+05
32	318.1831	C19 H26 O4	8.59	(9S.10S)-trans-Cannabitriol-C3/(9R.10R)-trans-cannabitriol-C3	4.9E+05
33	332.2353	C21 H32 O3	8.59	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	2.5E+06
34	316.2389	C21 H32 O2	8.81	Cannabinerol/abnormal cannabigerol/hexahydrocannabinol	2.2E+07
35	342.2193	C22 H30 O3	8.84	Ferruginene C/2-formyl-delta9-trans-tetrahydrocannabinol	1.1E+06
36	332.2354	C21 H32 O3	8.84	rac-6'-Epoxycannabigerol (2'S*. 3'R*)/rac-6'-epoxycannabigerol (2'R*. 3'R*)/(-)-7-Hydroxycannabichromane	6.3E+06
37	312.2096	C21 H28 O2	8.91	7.8-Dihydrocannabinol	3.3E+06
38	372.2306	C23 H32 O4	8.91	(+/-)-4-Acetoxycannabichromene/acetyl cannabigeroquinol/7.8-dehydro-10-O-ethylcannabitriol/	3.2E+06
39	316.2388	C21 H32 O2	9.20	Cannabinerol/abnormal cannabigerol/hexahydrocannabinol	5.2E+06
40	268.1465	C18 H20 O2	9.31	Cannabinol-C2/56a/63a	3.9E+05
41	314.2253	C21 H30 O2	9.38	(1aS.3aR.8bR.8cR)-Cannabicyclol/cannabicitran/(-)-delta9-cis-(6aS.10aR)-delta9-tetrahydrocannabinol/(âˆ’)-delta7 -trans-(1R. 3R. 6R)-isotetrahydrocannabinol-C5/ cannabigeroquinone	1.4E+07
42	374.2460	C23 H34 O4	9.39	(-)-(9R.10R)-trans-10-O-Ethylcannabitriol/5-acetyl-4-hydroxycannabigerol/acetyl abnormal cannabigeroquinol/cannabigerolic acid monomethylether	7.7E+06
43	342.2200	C22 H30 O3	9.47	Ferruginene C/2-formyl-delta9-trans-tetrahydrocannabinol	1.4E+07
44	328.2041	C21 H28 O3	9.49	Cannabichromanone-D/cannabicoumaronone/10-oxo-delta-6a(10a)-tetrahydrocannabinol/8-Oxo-delta9-trans-tetrahydrocannabinol/9.10-Anhydrocannabitriol/anhydrocannabimovone/Cannabidiol Hydroxyquinone	3.3E+07

Table III - continuation: Overview of compounds detected in ESI- (corresponding with the Figure 4)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Peak area[e]
45	312.2091	C21 H28 O2	10.05	7.8-Dihydrocannabinol	1.9E+06
46	372.2303	C23 H32 O4	10.05	(+/-)-4-Acetoxycannabichromene/acetyl cannabigeroquinol/7.8-dehydro-10-O-ethylcannabitriol/	1.8E+06
47	380.2714	C26 H36 O2	10.21	O-Penthylcannabinol	1.1E+06
48	384.3029	C26 H40 O2	10.23	Sesquicannabigerol/O-propylcannabidiol/O-pentyl-delta9-trans-tetrahydrocannabinol	6.1E+06
49	384.3025	C26 H40 O2	10.76	Sesquicannabigerol/O-propylcannabidiol/O-pentyl-delta9-trans-tetrahydrocannabinol	4.9E+06
50	686.4188	C43 H58 O7	11.10	cannabidiolic acid tetrahydrocannabitriol ester	7.2E+06
51	358.2144	C22 H30 O4	11.68	Ferruginene A/ferruginene B/delta-9-tetrahydrocannabinolic acid B/cannabichromenic acid/cannabicyclolic acid/delta-8-tetrahydrocannabinolic acid	9.5E+05
52	640.4493	C43 H60 O4	12.52	Cannabisol	5.8E+07
53	686.4189	C43 H58 O7	12.92	cannabidiolic acid tetrahydrocannabitriol ester	8.5E+05
54	494.3402	C32 H46 O4	13.33	beta-Fenchyl delta 9 - tetrahydrocannabinolate/epi-bornyl delta 9 - tetrahydrocannabinolate/alpha -terpenyl delta 9 -tetrahydrocannabinolate/4-terpenyl delta 9 - tetrahydrocannabinolate/bornyl delta 9 - tetrahydrocannabinolate/alpha -fenchyl delta 9 - tetrahydrocannabinolate	1.9E+07
55	640.4490	C43 H60 O4	13.41	Cannabisol	4.9E+06
56	640.4484	C43 H60 O4	13.56	Cannabisol	2.4E+06

[a] *Mass error compared the theoretical exact mass < 5 ppm*
[b] *Match of isotopic pattern confirmed*
[c] *Retention time*
[d] *Tentative identification of compounds based on available scientific articles (Hanuš et al. 2016, Mechoulam 2002); possible identities are separated by the symbol "/"*
[e] *Peak area related to the undiluted sample prepared according to stated procedure*

B) Targeted screening of other (non-cannabinoid) biologically active compounds

In the tested samples, the following amounts of other (non-cannabinoid) biologically active compounds (**Table IV**) characterized by unique combination of exact mass and retention time RT were detected.

Table IV: Number of other biologically active compounds detected in the sample in ESI+ and ESI-

Sample name	Number of other biologically active compounds detected in ESI+	Number of other biologically active compounds detected in ESI-
ML 2340/21	1	25

Extracted ion chromatograms (XIC) of the detected compounds in both ionization modes (ESI+, ESI-) are documented on **Figures 5 - 6**. All detected non-cannabinoid biologically active compounds (tentative identification) are also summarized in **Table V** (ESI+) and **Table VI** (ESI-). Considering the possible existence of isomeric forms, one detected compound may have several identities.

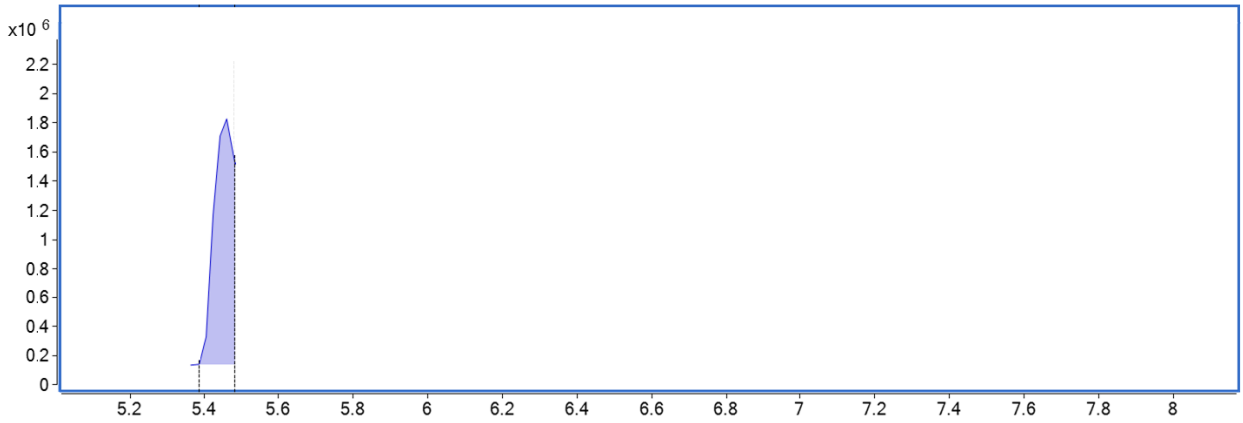

Figure 5: ML 2340/21 - UHPLC-HRMS/MS extracted ion chromatogram of 1 compound with unique combination of exact mass and RT detected in ESI+ (obtained for undiluted sample)

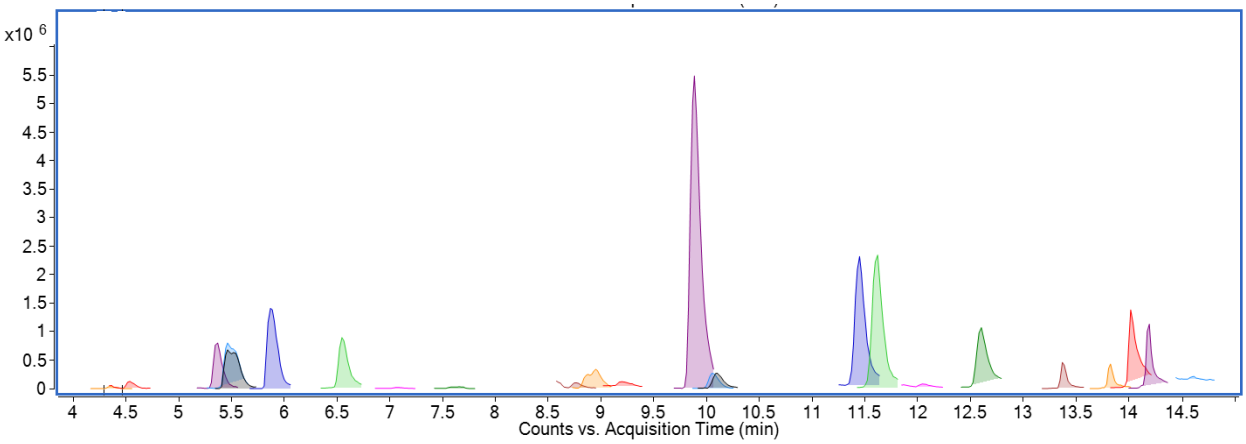

Figure 6: ML 2340/21 - UHPLC-HRMS/MS extracted ion chromatogram of 25 compounds with unique combination of exact mass and RT detected in ESI- (obtained for undiluted sample)

Table V: Overview of compounds detected in ESI+ (corresponding with the Figure 5)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Compound group	Peak area[e]
1	302.1139	C17 H18 O5	5.45	4.5-dihydroxy-2.5-dihydroxy-2.3.6-trimethoxy-9.10-dihydrophenanthrene	new non-cannabinoid constituent[f]	3.0E+06

[a] *Mass error compared the theoretical exact mass < 5 ppm*
[b] *Match of isotopic pattern confirmed*
[c] *Retention time*
[d] *Possible identities are separated by the symbol "/"*
[e] *Peak area related to the undiluted sample prepared according to stated procedure*
[f] *Mohamed M. Radwan et al. 2008*

Table VI: Overview of compounds detected in ESI- (corresponding with the Figure 6)

Cmp. number	Measured exact mass[a] (neutral, monoisotopical)	Estimated elemental formula[b]	RT[c] (min)	Tentative identity (name of possible compounds)[d]	Compound group	Peak area[e]
1	246.1256	C15 H18 O3	4.36	cannabispiran / isocannabispiran	phenol	2.4E+05
2	242.0948	C15 H14 O3	4.53	isocannabispiradienone / cannithrene-1	phenol	7.0E+05
3	246.1258	C15 H18 O3	5.37	cannabispiran / isocannabispiran	phenol	4.8E+06
4	242.0947	C15 H14 O3	5.46	isocannabispiradienone / cannithrene-1	phenol	6.9E+06
5	302.1152	C17 H18 O5	5.53	4.5-dihydroxy-2.5-dihydroxy-2.3.6-trimethoxy-9.10-dihydrophenanthrene	new non-cannabinoid constituent[f]	6.6E+06
6	272.1050	C16 H16 O4	5.87	cannithrene-2	phenol	1.6E+07
7	144.1154	C8 H16 O2	6.54	caprylic acid	fatty acid	6.5E+08
8	158.1306	C9 H18 O2	7.05	pelargonic acid	fatty acid	1.4E+06
9	172.1463	C10 H20 O2	7.61	capric acid	fatty acid	2.5E+05
10	256.2400	C16 H32 O2	8.55	palmitic acid	fatty acid	3.1E+07
11	200.1777	C12 H24 O2	8.76	lauric acid	fatty acid	8.1E+05
12	372.2306	C23 H32 O4	8.91	5-acetoxy-6-geranyl-3-n-pentyl-1.4-benzoquinone	new non-cannabinoid constituent[f]	3.2E+06
13	256.2402	C16 H32 O2	9.20	palmitic acid	fatty acid	1.2E+06
14	278.2250	C18 H30 O2	9.88	alpha-linolenic acid / gamma-linolenic acid / isolinolenic acid	fatty acid	3.9E+07
15	372.2303	C23 H32 O4	10.05	5-acetoxy-6-geranyl-3-n-pentyl-1.4-benzoquinone	new non-cannabinoid constituent[f]	1.8E+06
16	228.2091	C14 H28 O2	10.10	myristic acid	fatty acid	1.9E+06
17	256.2406	C16 H32 O2	11.45	palmitic acid	fatty acid	7.9E+08
18	282.2565	C18 H34 O2	11.61	cis-vaccenic acid / oleic acid	fatty acid	1.7E+07
19	270.2559	C17 H34 O2	12.04	margaric acid	fatty acid	4.0E+05
20	284.2719	C18 H36 O2	12.60	stearic acid	fatty acid	6.3E+08
21	312.3029	C20 H40 O2	13.37	arachidic acid / isoarachidic acid	fatty acid	1.8E+06
22	340.3341	C22 H44 O2	13.82	behenic acid	fatty acid	1.9E+06
23	414.3861	C29 H50 O	14.02	beta-sitosterol	phytosterol	7.9E+07
24	368.3653	C24 H48 O2	14.18	lignoceric acid	fatty acid	1.8E+07
25	284.2715	C18 H36 O2	14.62	stearic acid	fatty acid	3.6E+05

[a] *Mass error compared the theoretical exact mass < 5 ppm*
[b] *Match of isotopic pattern confirmed*
[c] *Retention time*
[d] *Possible identities are separated by the symbol "/"*
[e] *Peak area related to the undiluted sample prepared according to stated procedure*
[f] *Mohamed M. Radwan et al. 2008*

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1. Hanus, L.O., et al., *Phytocannabinoids: a unified critical inventory*. Natural Product Reports, 2016. **33**(12): p. 1357-1392.
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End of the appendix

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