

APPLICATION NOTE

Analysis of Extractable & Leachable Compounds from Drug Formulations using Perkin Elmer GC-MS Systems

The purpose of Extractable & Leachables analysis is to identify and quantify possible harmful compounds that can migrate (Leachables) from primary packaging systems and medical devices into pharmaceuticals and other finished products. The E&L analysis is mandatory to be carried out based on a toxicological risk assessment following regulatory requirements.

Identification of potentially toxic chemicals leaching from a great variety of polymers and plastics destined for pharmaceutical packaging products has received an important attention, but remains a quite challenging analysis for testing laboratories.

Most common additives used in pharmaceutical primary packaging systems are:

- Plasticizers;
- Flame retardants;
- Impact modifiers;
- Antioxidants;
- Antimicrobials;
- UV Stabilizers;
- Colorants;
- Lubricants and processing aids.

Extractables and Leachables studies using gas chromatography-mass spectrometry (GC/MS) are designed to detect volatile and some semi-volatile compounds from medical devices and closure systems. For the semi-volatiles analysis, direct liquid injection is preferred, while for the volatiles the headspace technique is most advantageous.



Extractables testing involves exposing the primary packaging system and its components to organic solvents / water at different pH, high temperatures, or an extended period of time to simulate a leachable profile. This is called the worst-case scenario and it must give information (qualitative and quantitative) about all the compounds that can leach into drug formulation.

In Figure 1. a LDPE dropper system was used in an Extractable study with organic solvents (IPA and n-Hexane) and water at acidic and alkaline pH in a reflux system for 6 hours. The chromatograms were recorded with a Perkin Elmer GC-MS Clarus 680 / Clarus SQ 8T.

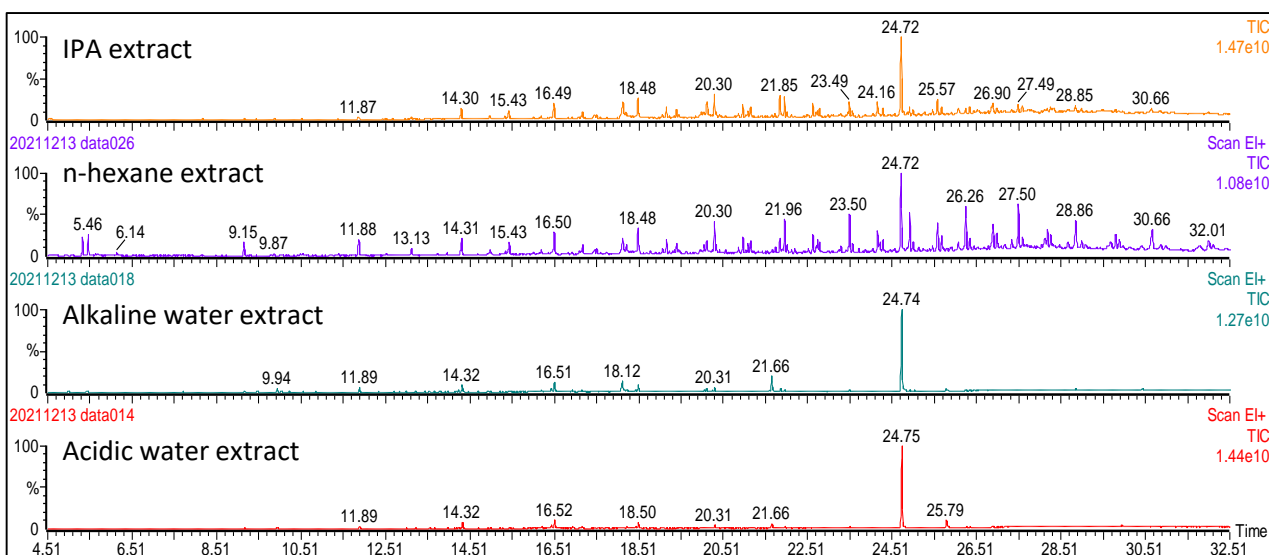


Figure 1. GC-MS total ion chromatogram obtained in the Extractables study of a LDPE system with IPA, n-hexane and water as extraction solvents.

Table 1. Compounds detected in LDPE system extracted with IPA and n-hexane using GC-MS (liquid injection and Headspace)

#	Name	CAS	Match	Common uses	#	Name	CAS	Match	Common uses
1	Dodecane	112-40-3	936	Plasticizer	16	Pentacosane	629-99-2	889	LDPE degradation product
2	Tetradecane	629-59-4	942	Plasticizer	17	1-Cyclopentyleicosane	-	885	
3	Hexadecane	544-76-3	918	Plasticizer	18	Octacosane	630-02-4	913	LDPE degradation product
4	Heptadecane	629-78-7	924	Plasticizer	19	Octacosyl pentyl ether	-	875	
5	1-Octadecanol	112-92-5	878	Lubricant	20	Tetratetracontane	7098-22-8	909	LDPE degradation product
6	Octadecane	593-45-3	918	Plasticizer	21	Pentyl triacontyl ether	-	858	
7	Eicosane	112-95-8	937	LDPE degradation product	22	Hentriacontane	630-04-6	911	LDPE degradation product
8	1-Eicosanol	629-96-9	883		23	Dotriacontyl methyl ether	-	831	
9	3-Heptadecanone	84534-29-2	753		24	Tetratetracontane	7098-22-8	901	LDPE degradation product
10	Hexadecanamide	629-54-9	860		25	Tetratriacontyl pentafluoropropionate	-	842	
11	Heneicosane	629-94-7	907	LDPE degradation product	26	Hentriacontane	630-04-6	899	LDPE degradation product
12	1-Docosanol, methyl ether	-	878		27	Hexatriacontyl pentafluoropropionate	-	849	
13	3-Pentadecanone	18787-66-1	749						
14	Octadecanamide	124-26-5	829						
15	n-Tetracosanol-1	506-51-4	893						

Extractables

Leachables studies are performed on the actual drug formulation under normal or accelerated storage conditions.

Leachable compounds are typically part of the extractables because of the direct contact of drug formulation with the packaging material.

Leachables
Volatiles

For the analysis of volatile leachables, we developed, optimized and validated a Headspace – GC – MS method able to identify and quantify the possible harmful compounds at low limits of detection.

Leachables
Semi-volatiles

The quantification of leachables is based on the relative response factor approach calculated from a calibration solution that contains ethanol, methyl-isobutyl-ketone, toluene, 1-propanol, n-decane. 1,4-Dioxane is the internal standard.

Results reporting

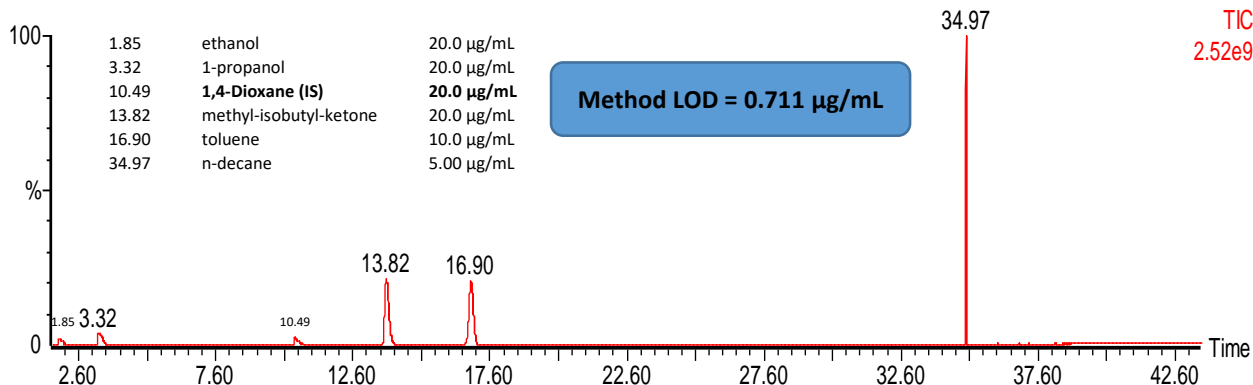


Table 2. HS-GC-MS analytical parameters used for E&L studies

Perkin Elmer TurboMatrix S40	
Oven temperature (°C)	80
Needle temperature (°C)	120
Loop temperature (°C)	125
Incubation time (min)	60
Pressurization time (min)	3.0
Vial pressure (psi)	20.0

Perkin Elmer GC Clarus 680	
Column	Elite-Volatile 30m x 0.25mm x 1.40µm
Injector temperature (°C)	200
Injection time (min)	0.10
Injection mode	split injection @ split ratio 20:1
Carrier gas, Mode/Flow	He, constant flow @ 2.00 mL/min

GC oven temperature program	
Temperature 1 (°C)	35
Hold time 1 (min)	7
Temperature 2 (°C)	40
Rate (°C/min)	1
Hold time 2 (min)	15
Temperature 3 (°C)	100
Rate (°C/min)	10
Temperature 4 (°C)	240
Rate (°C/min)	25
Hold time 3 (min)	5

Perkin Elmer MS SQ8T	
Inlet temperature (°C)	180
Source temperature (°C)	150
Acquisition mode	EI+ @ 70eV, scan range 25-200

Figure 2. System Suitability Solution for Headspace-GC-MS

Extractables

For the analysis of semi-volatile leachables, a direct liquid injection GC – MS method is used to identify and quantify the compounds at sub-ppm limits of detection.

The quantification of leachables is based on the relative response factor approach calculated from a calibration solution prepared from Grob Test Mix (Restek, P/N 35000). Irganox 415 is the internal standard.

For System Suitability the acceptance criteria is:

- Resolution between n-undecane and n-nonanal must be not less than 1.0
- Signal-to-noise ratio for 1-octanol peak must be not less than 3:1

Leachables
Volatiles

Leachables
Semi-volatiles

Results
reporting

Table 3. Liquid injection- GC-MS analytical parameters used for E&L studies

Perkin Elmer Liquid Autosampler	
Syringe capacity volume (µL)	5.0
Injection speed	Normal
Viscosity delay (s)	3
Pre-injection solvent wash	2
Pre-injection sample wash	1
Post-injection solvent wash	5

Perkin Elmer GC Clarus 680	
Column	Elite-5MS 30m x 0.25mm x 0.25 µm
Injector temperature (°C)	300
Injection volume (µL)	1.0
Injection mode	split injection @ split ratio 10:1
Carrier gas, Mode/Flow	He, constant flow @ 1.20 mL/min
GC oven temperature program	
Temperature 1 (°C)	50
Hold time 1 (min)	1
Temperature 2 (°C)	8
Rate (°C/min)	315
Hold time 2 (°C)	5

Perkin Elmer MS SQ8T	
Inlet temperature (°C)	315
Source temperature (°C)	230
Acquisition mode	EI+ @ 70eV, scan range 30 - 620

n-decane	14.06 µg/mL
1-octanol	18.01 µg/mL
n-undecane	14.54 µg/mL
n-nonanal	20.01 µg/mL
2,6-dimethylphenol	16.01 µg/mL
2,6-dimethylaniline	16.07 µg/mL
methyl decanoate	21.04 µg/mL
methyl undecanoate	21.06 µg/mL
methyl dodecanoate	20.53 µg/mL
Irganox 415 (IS)	40.00 µg/mL

Method LOD = 0.607 µg/mL

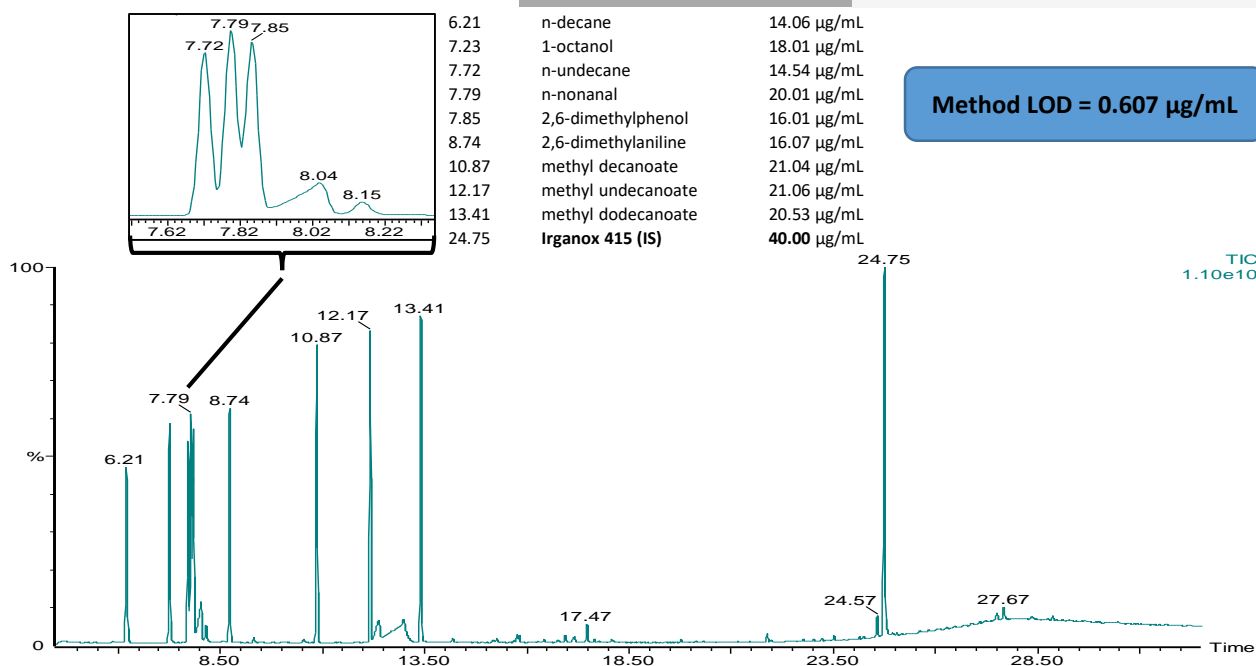


Figure 3. System Suitability Solution (prepared as 20x dilution from Grob Test Mix, RESTEK PN 35000)

Extractables

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Compound identification

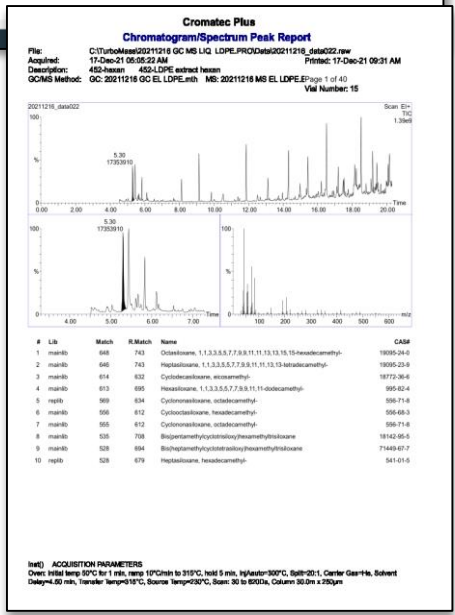
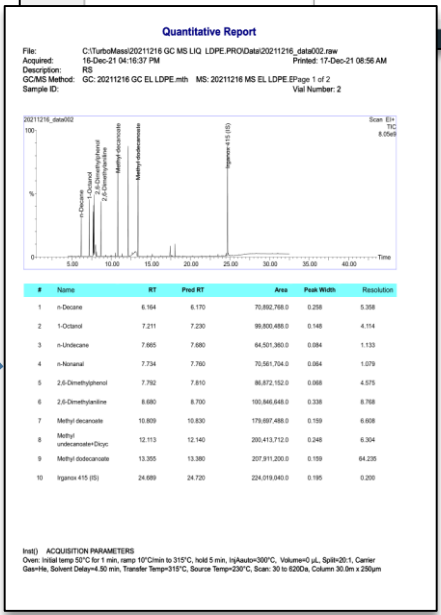
Compounds were characterized using the Perkin Elmer TurboMass software (version 6.1.2) and Communicue software to generate the reports.

Mass spectra of all compounds from all compounds were matched with the NIST Library 2.2 and Wiley9 Library. Compounds with a mass spectral match of at least 700 were considered, and the top match was used in the result reporting.

Perkin Elmer TurboMass Software (ver. 6.1.2)

Peak	Retention Time (min)	Compound Name	Library	Match Score	Abundance
1	20211216.000001	Blank DCM	20211216.000001.DCM	1	Blank
2	20211216.000002	Blank DCM	20211216.000002.DCM	1	Blank
3	20211216.000003	Soluble vitamins B5	20211216.000003.DCM	2	GC 76
4	20211216.000004	System Suitability 15.1	20211216.000004.DCM	3	GC 15.1
5	20211216.000005	System Suitability 15.2	20211216.000005.DCM	3	GC 15.2
6	20211216.000006	System Suitability 15.3	20211216.000006.DCM	3	GC 15.3
7	20211216.000007	System Suitability 15.4	20211216.000007.DCM	3	GC 15.4
8	20211216.000008	System Suitability 15.5	20211216.000008.DCM	3	GC 15.5
9	20211216.000009	System Suitability 15.6	20211216.000009.DCM	3	GC 15.6
10	20211216.000010	Blank DCM	20211216.000010.DCM	1	Blank
11	20211216.000011	System Suitability 15.6	20211216.000011.DCM	4	GC 15.6
12	20211216.000012	Matrix Extracted (Sample) 100%	20211216.000012.DCM	5	Analysis Matrix 100%
13	20211216.000013	Matrix Extracted (Sample) acid	20211216.000013.DCM	6	Analysis Matrix acid
14	20211216.000014	452-Heptan-4-ol	20211216.000014.DCM	7	Analysis 452-Heptan-4-ol
15	20211216.000015	452-Heptan-4-ol	20211216.000015.DCM	8	Analysis 452-Heptan-4-ol
16	20211216.000016	452-Heptan-4-ol	20211216.000016.DCM	9	Analysis 452-Heptan-4-ol
17	20211216.000017	452-Heptan-4-ol	20211216.000017.DCM	10	Analysis 452-Heptan-4-ol
18	20211216.000018	452-Heptan-4-ol	20211216.000018.DCM	11	Analysis 452-Heptan-4-ol
19	20211216.000019	452-Heptan-4-ol	20211216.000019.DCM	12	Analysis 452-Heptan-4-ol
20	20211216.000020	452-Heptan-4-ol	20211216.000020.DCM	13	Analysis 452-Heptan-4-ol
21	20211216.000021	Blank DCM	20211216.000021.DCM	1	Blank
22	20211216.000022	System Suitability 15.1	20211216.000022.DCM	3	GC 15-021
23	20211216.000023	System Suitability 15.2	20211216.000023.DCM	3	GC 15-022
24	20211216.000024	Blank DCM	20211216.000024.DCM	1	Analysis Blank
25	20211216.000025	Matrix Extracted (Sample)	20211216.000025.DCM	14	Analysis Matrix (Sample)
26	20211216.000026	452-Heptan-4-ol	20211216.000026.DCM	15	Analysis 452-Heptan-4-ol
27	20211216.000027	452-Heptan-4-ol	20211216.000027.DCM	16	Analysis 452-Heptan-4-ol
28	20211216.000028	452-Heptan-4-ol	20211216.000028.DCM	17	Analysis 452-Heptan-4-ol
29	20211216.000029	452-Heptan-4-ol	20211216.000029.DCM	18	Analysis 452-Heptan-4-ol
30	20211216.000030	452-Heptan-4-ol	20211216.000030.DCM	19	Analysis 452-Heptan-4-ol
31	20211216.000031	452-Heptan-4-ol	20211216.000031.DCM	20	Analysis 452-Heptan-4-ol
32	20211216.000032	452-Heptan-4-ol	20211216.000032.DCM	21	Analysis 452-Heptan-4-ol
33	20211216.000033	Blank DCM	20211216.000033.DCM	1	Blank
34	20211216.000034	System Suitability 15.1	20211216.000034.DCM	3	GC 15-023

Communicue™



System Suitability Report using Communicue – peak area + resolution

Qualitative Report using Communicue – NIST, Wiley libraries integrated