Case Study: Method Optimization and Extractables Characterization of a Peroxide Cured Rubber, Alan D. Hendricker, Andrea Deal, and Thomas N. Feinberg, Cardinal Health, Research Triangle Park, NC

Outline/Introduction

Extractables were characterized from a peroxide cured rubber by extraction in various solvents, via various methods followed by extract analysis by gas chromatography-mass spectrometry (GC-MS), liquid chromatography-mass spectrometry (LC-MS). Based on the generated profiles, peaks were identified to the extent feasible, compared with supplier provided information and an optimal extraction technique was chosen (methylene chloride reflux). The methods were demonstrated to be sensitive for other target compounds, including compounds requiring special considerations.

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Choosing Extraction Solvents and Techniques

Extraction solvents of hexane, isopropanol and methylene chloride were chosen based on the working committee guidelines/recommendations:

They represent a range of polarities, and therefore potential solubilizing properties.

They represent a range of boiling points.

They are relatively non-reactive chemically.

They are easily handled in a typical analytical laboratory setting.

They are readily available in high purity.

Extraction techniques of reflux, sonication and Soxhlet were also based on working committee guidelines/recommendations:

In the experience of Working Group members, these three techniques are, and have been, in common use in the industry for extractables studies and testing.

Each of these techniques has a long history of varied and effective use in the scientific literature.

All three extraction techniques employ equipment which is routinely available in a typical analytical laboratory.

Other extraction techniques are possible, including maceration, supercritical fluid extraction, Dionex accelerated solvent extraction, etc. Other solvents are possible and should be based on a variety of considerations, including compatibility with materials, analytical techniques, reproducibility, safety, etc.

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Extraction Procedures and Sample Preparation

The peroxide cured elastomer sample was provided by West Pharmaceutical Services in cut 4 3/8 inch squares. Samples were cut into smaller pieces using scissors prior to extraction.

The peroxide cured elastomer was extracted in three different solvents: methylene chloride, isopropanol and hexane. Extractions were performed by sonication in each solvent for 30 minutes and 2 hours. Extractions were also performed by reflux in each solvent for 4 hours and Soxhlet extraction for 16 hours. Extraction blanks were prepared in a similar manner in all cases. but without sample addition.

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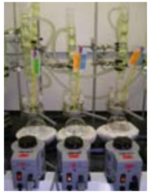

Picture of Peroxide Cured Elastomer

Elastomer Formulation

•Ingredient	%
•Imsil A25 (silicone dioxide)	24.01
•Mistron Cyprubond (magnesium silicate)	19.21
•Bromobutyl 2030	38.42
•VISTALON 404 (ethylene propylene copolymer)	9.61
•White oil 2	1.44
•420 blue MB	0.12
•Titanium dioxide	1.68
•Paraffin	0.96
•Magnesium oxide	0.60
•Stearic acid	0.48
•Polyethylene wax	1.44
•P-800	2.03

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GC-MS Analytical Parameters

GC

Instrument: Hewlett-Packard 6890

Injection Mode: Hewlett-Packard splitless

Injection Volume: 1 µL

Injector Program: 280 °C

Purge Valve: On at 1.00 min., off initially

Column: Restek Rtx-1, 30 m x 0.25 mm (0.1 mm film) or equivalent

Oven Temperature: 40 °C for 1 min., heated at 10 °C/min. to 300 °C and hold for 10 min.

Constant flow (helium) at 1 mL/min.

Transfer Line: 280 °C

MS

Instrument: Hewlett-Packard 5972 or 5973 MSD

Ionization Mode: EI (electron ionization)

Scan Mode: Scanning; m/z 50-650

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LC-MS Analytical Parameters

LC

HPLC Instrument: Hewlett-Packard 1050

Injection Volume: 50 µL

UV Wavelength: 280 nm

Flow Rate: 1 mL/min

Column: Alltech Alltima C18, 4.6 mm x 25 cm 5 µm particles

Mobile Phase: A – 75:25 acetonitrile/water
B – 50:50 acetonitrile/tetrahydrofuran

Gradient:

Time (minutes)	% A	% B
0	100	0
10	60	40
20	0	100
30	0	100
32	100	0
45	100	0

MS

MS Instrument: Micromass Platform II

Ionization Mode: APCI (Atmospheric Pressure Chemical Ionization) both APCI+ and APCI-

Scan Range: m/z 50-1350

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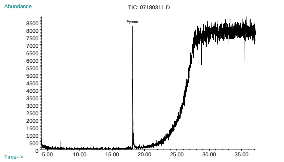
Method Sensitivity (GC-MS) 1 ng Pyrene Injected (1 µg/mL standard solution)



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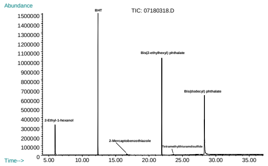
Method Standard Chromatography Example (GC-MS) 50 ng of each Standard Injected



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Method Reproducibility Standard (GC-MS) of Various Standards

	100 ng injection	1 ng injection	50 ng injection	50 ng injection
Trial	**Stearic Acid**	**Pyrene**	**2-ethylhexanol**	**BHT**
	Area	Area	Area	Area
1	10756261	214440	8514431	29799955
2	11722863	203118	9526329	32983684
3	12936169	169879	10156015	33074210
4	11415222	190669	10164135	33748099
5	12537378	188110	10305556	32913801
%RSD	7.36	8.68	7.65	4.76
	50 ng injection	50 ng injection	50 ng injection	50 ng injection
Trial	**2-mercaptobenzothiazole**	**dioctyl phthalate**	**tetramethyl disulfide**	**didodecyl phthalate**
	Area	Area	Area	Area
1	1678118	27684976	898422	21042469
2	2235219	29938620	902084	22850265
3	2248637	30457087	924195	23225873
4	2590970	29467944	761762	23798223
5	2375607	30939982	719121	24421984
%RSD	15.18	4.22	11.13	5.54

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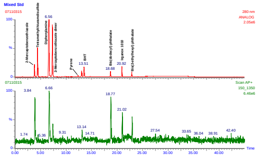
Example Chromatography, Mixed Standard (50 – 500 ng injected) by Positive Ion APCI LC-MS: UV Traces were used for Quantitation



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Method Reproducibility Standard (LC-MS) of Various Standards

	500 ng injected	250 ng injected	250 ng injected	250 ng injected	50 ng injected
Trial	**Stearic Acid**	**2-mercaptobenzothiazole**	**Tetramethylthiuram**	**Diphenylamine**	**Pyrene**
	Area	**Area**	**Area**	**Area**	**Area**
1	863974	42720	127048	358829	18712
2	820366	42909	127951	360338	18526
3	904421	42542	127070	359341	18729
4	772242	42393	125309	359586	19157
5	800600	42605	124055	358483	20266
%RSD	6.29	0.45	1.25	0.20	3.69
	250 ng injected	250 ng injected	250 ng injected	250 ng injected	
Trial	**BHT**	**dioctyl phthalate**	**Irganox 1010**	**didodecyl phthalate**	
	Area	**Area**	**Area**	**Area**	
1	45482	17386	30376	11956	
2	45617	17376	30296	11906	
3	45528	17373	30312	11928	
4	45802	17278	30301	11777	
5	45560	17346	30364	11845	
%RSD	0.27	0.25	0.12	0.60	

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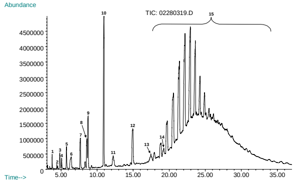
GC-MS TIC Peroxide Cured Elastomer, 4 Hour Methylene Chloride Reflux



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First Pass Identifications of Extractables (GC-MS Analysis) for Peroxide Cured Material

#	Retention Time	First Pass Identification
1	3.8	Unknown (possible peroxide fragment)
2	4.4	Unknown (possible peroxide fragment)
3	4.8	2-Methyl-3-bromo-2-butanol
4	5.1	P-800 Related Species (confirmed with authentic standard)
5	5.8	Unknown (possible peroxide fragment)
6	6.4	2,5-Dimethyl-2,5-hexanediol (peroxide fragment)
7	7.7	6-Bromohexanenitrile
8	8.6	P-800 Related Species (confirmed with authentic standard)
9	8.8	Unknown Base Polymer Species
10	11.0	2,5-dimethylhexane-2,5-dihydroperoxide
11	12.2	Brominated Base Polymer Oligomeric Species
12	14.9	Unknown Antioxidant Fragment
13	17.5	Hexadecanoic acid (Palmitic Acid)
14	19.3	Octadecanoic acid (Stearic Acid)
15	16-37	Aliphatic Saturated Hydrocarbons C22 – C40 (approx) originating from White Oil or Paraffin Component)

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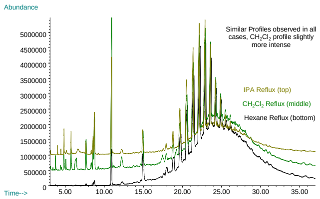
GC-MS TIC of Elastomer Reflux Extracts in Three Solvents



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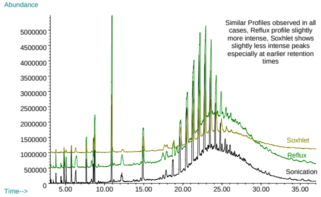
GC-MS TIC of Elastomer CH_2Cl_2 Extracts Via Three Methods



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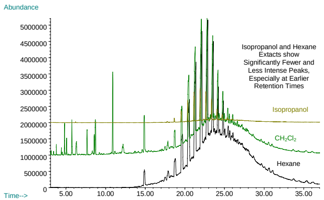
GC-MS TIC of Elastomer Sonication Extracts in Three Solvents



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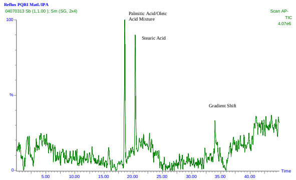
Negative Ion APCI TIC of Elastomer IPA Reflux Extract



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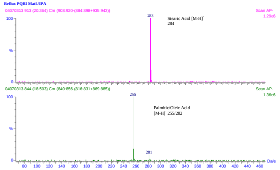
Negative Ion APCI Mass Spectra of Stearic and Palmitic/Oleic Acid Peaks (Source White Oil 2)



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Results and Conclusions

• Both LC-MS and GC-MS methods could detect a variety of target extractables at relatively low levels, including pyrene, a target with special considerations. Reproducibility of methods was appropriate for trace level and mass spectrometric methods chosen.

•Chromatographic separation of targets was appropriate.

•Methylene chloride reflux of the elastomer showed the most extractables and peaks of the greatest intensities and is may be the best method for detection of targets. However, degradants may occur which are not appropriate if conditions are too harsh.

•Appropriate studies should be done in which maximum amounts of extractables are achieved. This should be compared with known materials composition, if available.

•In this study GC-MS methodology showed many more extractables than LC-MS. Peaks known to be present in elastomer formulation can only be detected by GC methods (no chromophores).

•Test article profile in this study is complex. Difficult decisions must be made to determine appropriate targets and a method which is validatable for materials control.

•Several elastomer formulation components were not observed (via this methodology). Additional orthogonal techniques could be necessary for materials control (e.g. ICP-MS for control of Silicon, Magnesium, Titanium; Ion Chromatography for Bromide/Bromate).

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