GCxGC-TOFMS Utilized for Broad Spectrum Analyses of Endocrine Disruptor Compounds (EDCs)

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Endocrine Disruptors

- Endocrine disruptors: chemicals with the potential to interfere with the function of endocrine systems

- Long Term Effects:
  - reduced fertility
  - male and female reproductive tract abnormalities
  - skewed male/female sex ratios
  - brain and behavior problems
  - Impaired immune functions
  - various cancers (*i.e*. disinfection by-products linked to bladder cancer)

Reference: http://www.epa.gov/endocrine/

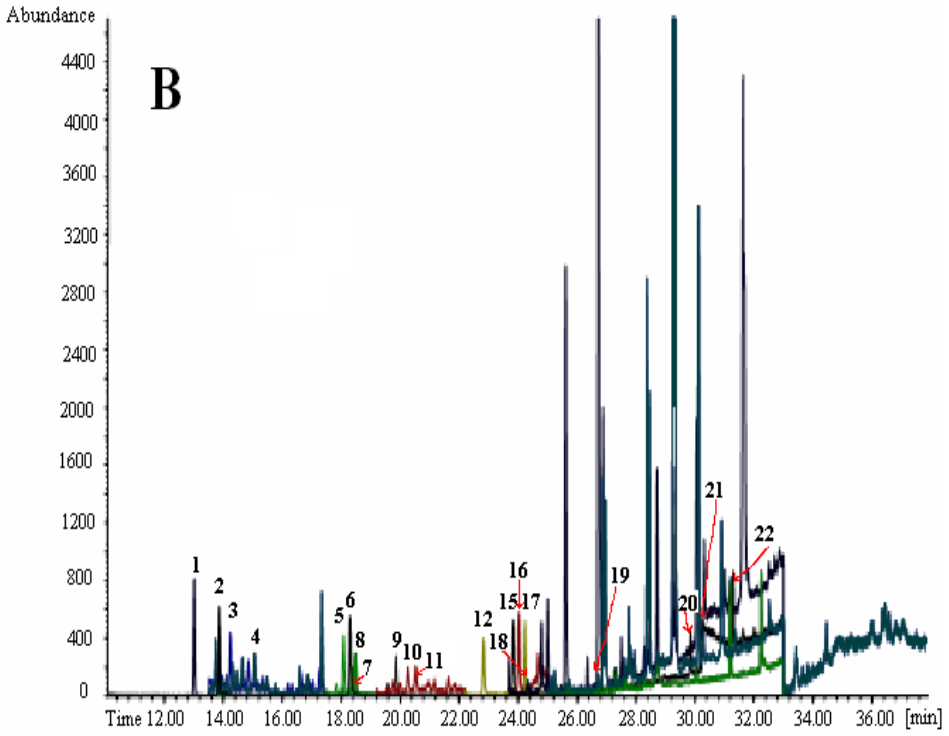
TEDX (The Endocrine Disruption Exchange, Inc.) LIST

endrin		72-20-8	H01345	1985	Abalis IM, Eldefrawi ME, Eldefrawi AT. 1985. High-affinity stereospecific binding of cyclodiene insecticides and gamma-hexachlorocyclohexane to gamma-aminobutyric acid receptors of rat brain. Pesticide Biochemistry & Physiology 24(1):95-102.
endrin		72-20-8	H13732	2004	Kojima H, Katsura E, Takeuchi S, Niiyama K, Kobayashi K. 2004. Screening for estrogen and androgen receptor activities in 200 pesticides by in vitro reporter gene assays using Chinese hamster ovary cells. Environ Health Perspect 112(5):524-531.
environmental tobacco smoke		n/a	H17509	2006	Slotkin TA, Pinkerton KE, Seidler FJ. 2006. Perinatal environmental tobacco smoke exposure in rhesus monkeys: Critical periods and regional selectivity for effects on brain cell development and lipid peroxidation. Environ Health Perspect 114(1):34-39.
epichlorohydrin	1-chloro-2,3-epoxypropane	106-89-8	H22563	1974	Cooper ERA, Jones AR, Jackson H. 1974. Effects of alpha-chlorohydrin and related compounds on the reproductive organs and fertility of the male rat. J Reprod Fertil 38(2):379-386.
epichlorohydrin	1-chloro-2,3-epoxypropane	106-89-8	H22614	1983	John JA, Quast JF, Murray FJ, Calhoun LG, Staples RE. 1983. Inhalation toxicity of epichlorohydrin: effects on fertility in rats and rabbits. Toxicol A[illegible] 415-423.
epichlorohydrin	1-chloro-2,3-epoxypropane	106-89-8	H08226	1983	Kluwe WM, Gupta BN, Lamb JC 4th. 1983. The comparative effects of 1,2-dibromo-3-chloropropane (DBCP) and its metabolites, 3-chloro-1,2-propaneoxide (epichlorohydrin), 3-chloro-1,2- propanediol (alphachlorohydrin), and oxalic acid, on the urogenital system of male rats. Toxicol Appl Pharmacol 70(1):67-86.
epichlorohydrin	1-chloro-2,3-epoxypropane	106-89-8	H08259	1989	Toth GP, Zenick H, Smith MK. 1989. Effects of epichlorohydrin on male and female reproduction in Long- Evans rats. Fundam Appl Toxicol 13(1):16-25.
epichlorohydrin	1-chloro-2,3-epoxypropane	106-89-8	H08256	1990	Slott VL, Suarez JD, Simmons JE, Perreault SD. 1990. Acute inhalation exposure to epichlorohydrin transiently decreases rat sperm velocity. Fundam Appl Toxicol 15(3):597-606.
EPN	ethyl p-nitrophenyl benzenethionophosphonate	2104-64-5	H13732	2004	Kojima H, Katsura E, Takeuchi S, Niiyama K, Kobayashi K. 2004. Screening for estrogen and androgen receptor activities in 200 pesticides by in vitro reporter gene assays using Chinese hamster ovary cells. Environ Health Perspect 112(5):524-531.
epofenonane		57342-02-6	W14671	2005	Oda S, Tatarazako N, Watanabe H, Morita M, Iguchi T. 2005. Production of male neonates in Daphnia magna (Cladocera, Crustacea) exposed to juvenile hormones and their analogs. Chemosphere 61(8):1168-1174.
epoxiconazole		133855-98-8 (formerly 106325-08-0)	H19711	2006	Trosken ER, Adamska M, Arand M, Zarn JA, Patten C, Volkel W, Lutz WK. 2006. Comparison of lanosterol-14 alpha-demethylase (CYP51) of human and Candida albicans for inhibition by different antifungal azoles. Toxicology 228(1):24-32.

Visit the TEDX website at http://www.endocrinedisruption.com

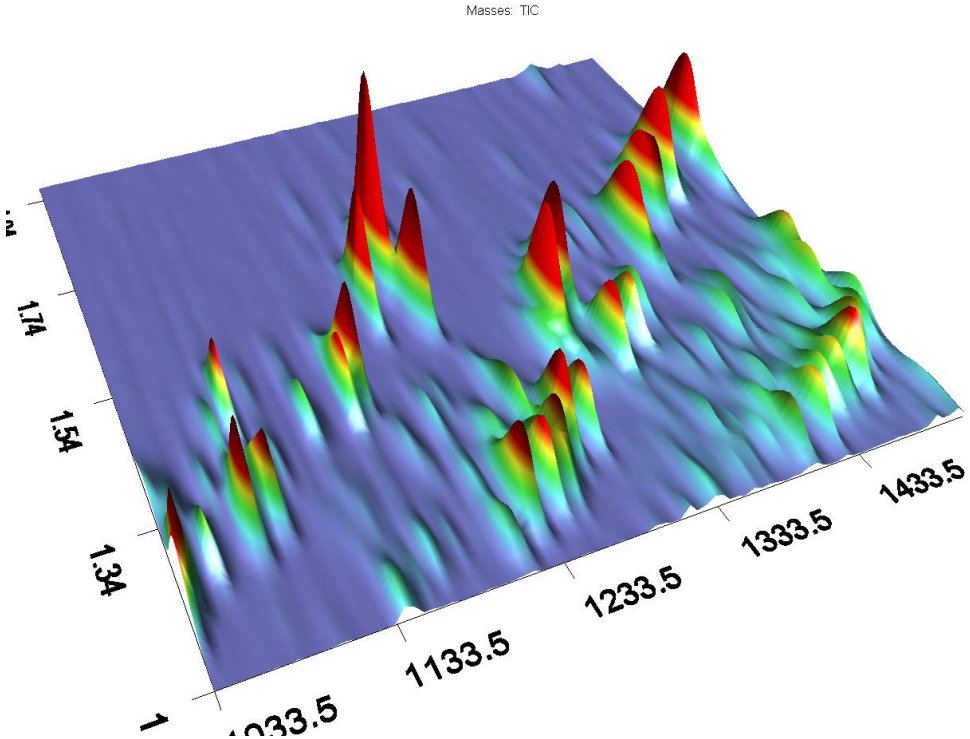
Analyses of EDCs
Targeted or Untargeted??

- LC/MS
- LC/MS/MS
- GC/MS
- GC/MS/MS
- GC-ToF
  - GC-ToF with GCxGC



Comprehensive GCxGC with LECO Pegasus 4D

Pegasus® 4D and HT

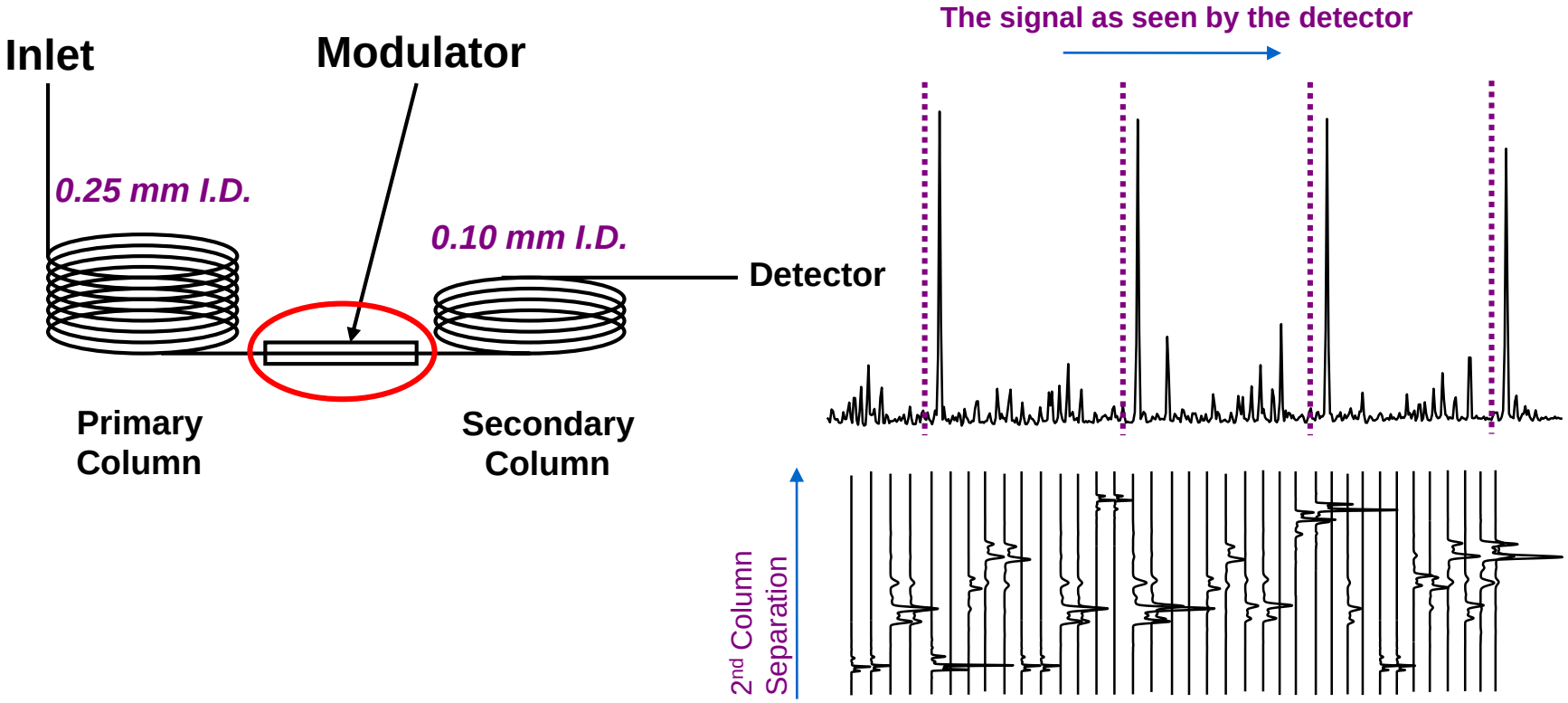


What is GCxGC?

Multi-Dimensional Gas Chromatography

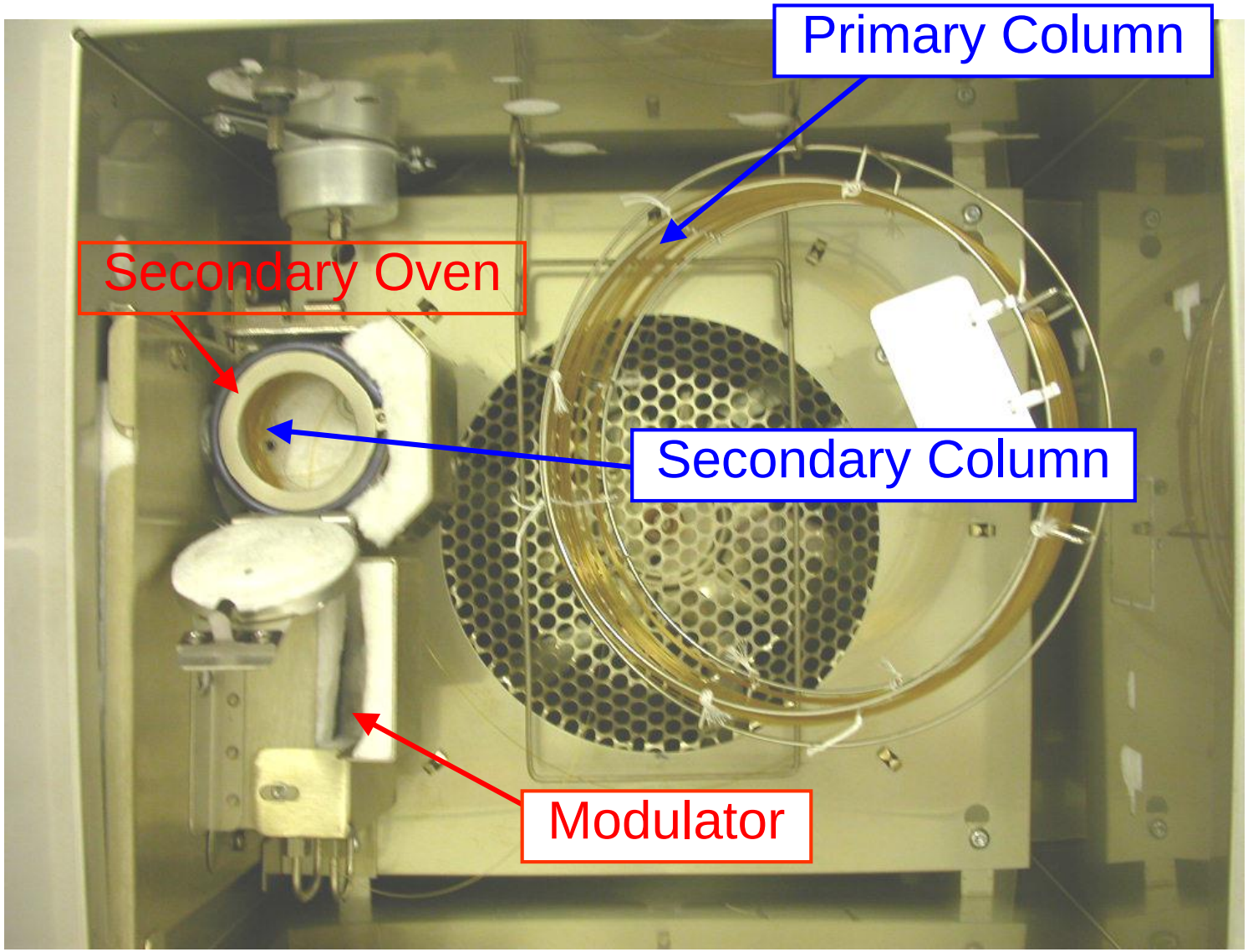
- GCxGC (Comprehensive Two-dimensional Gas Chromatography)
  - Comprehensive
    - » All material that enters the 1st dimension column (with stationery phase “A”) passes through the 2nd dimension column (with stationery phase “B”) to the same detector
  - Uses a “Modulator” to partition 1st column effluent as discrete plugs onto the 2nd dimension column

The GCxGC Process

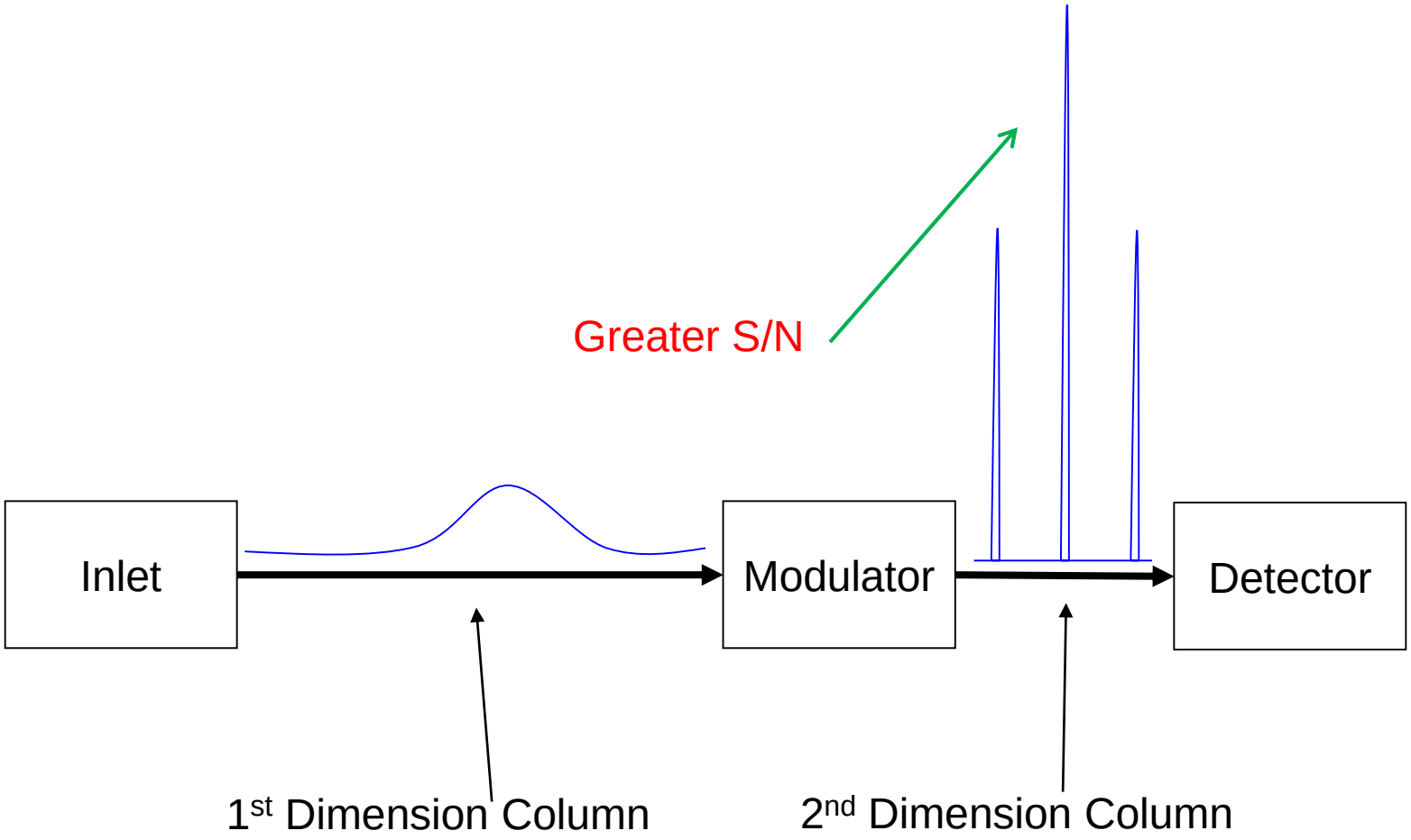


The effluent from the primary column is focused and segmented by the modulator into a discrete “plug”. Each plug is then injected onto the secondary column by the modulator, where it is separated. The GCxGC process is a series of independent second column separations.

LECO's GCxGC


Primary Column
Secondary Oven
Secondary Column
Modulator

Simplified GCxGC Flow

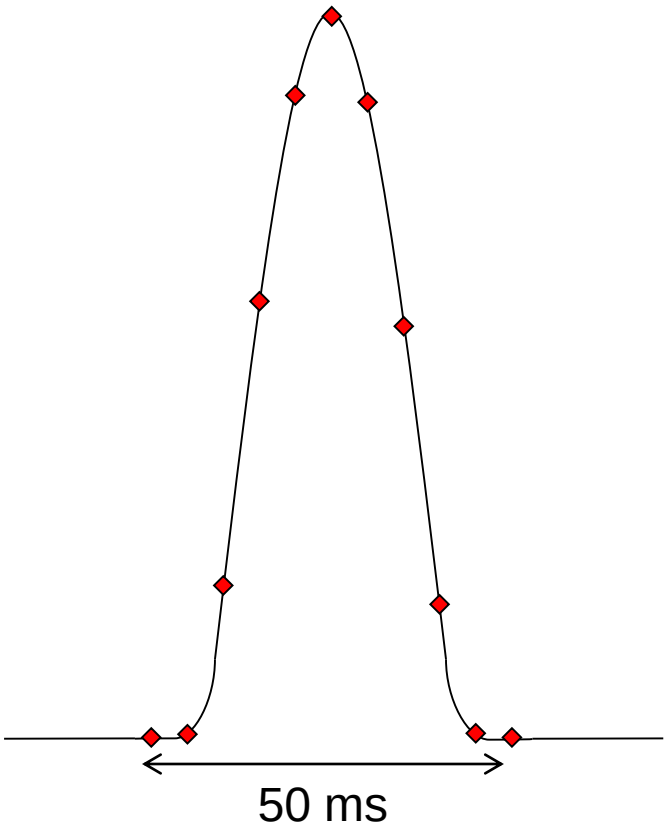


LECO GC-Time of Flight Mass Spec

Pegasus® 4D

Detector Requirements for GCxGC

Quantitation requires a minimum of 10 data points across a peak in order to define it



50 ms peak width at base

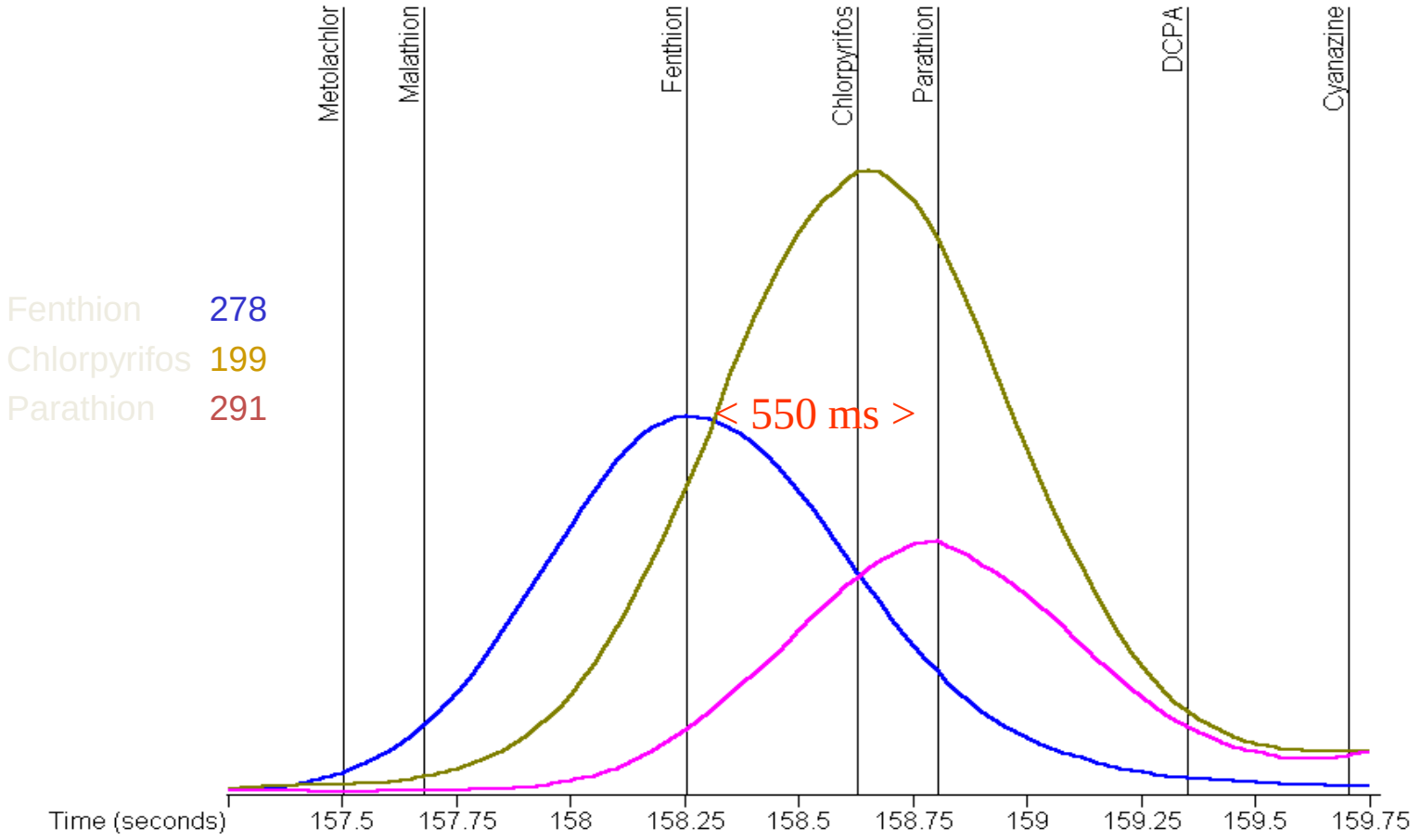
Need 10 data points / peak

5 ms between data points

Minimum Required Sampling Rate:

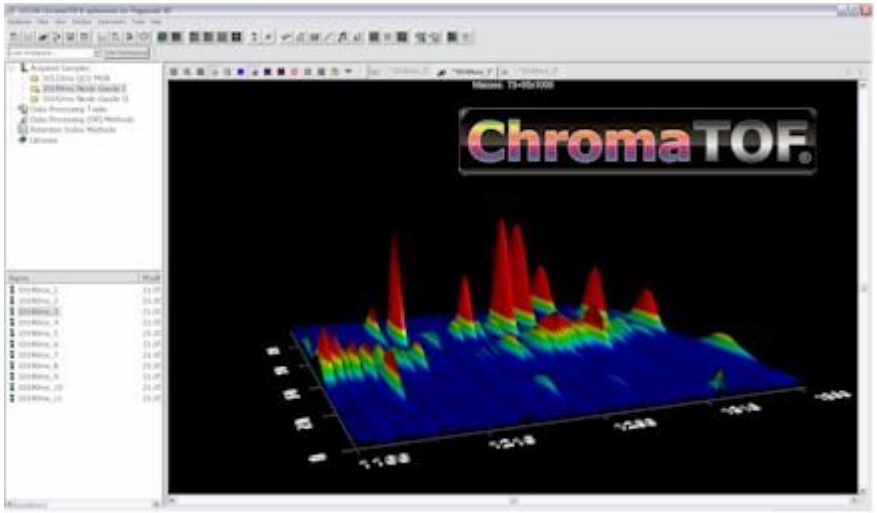
200 Hz for a 50 ms wide peak

Coelution of 3 Pesticides

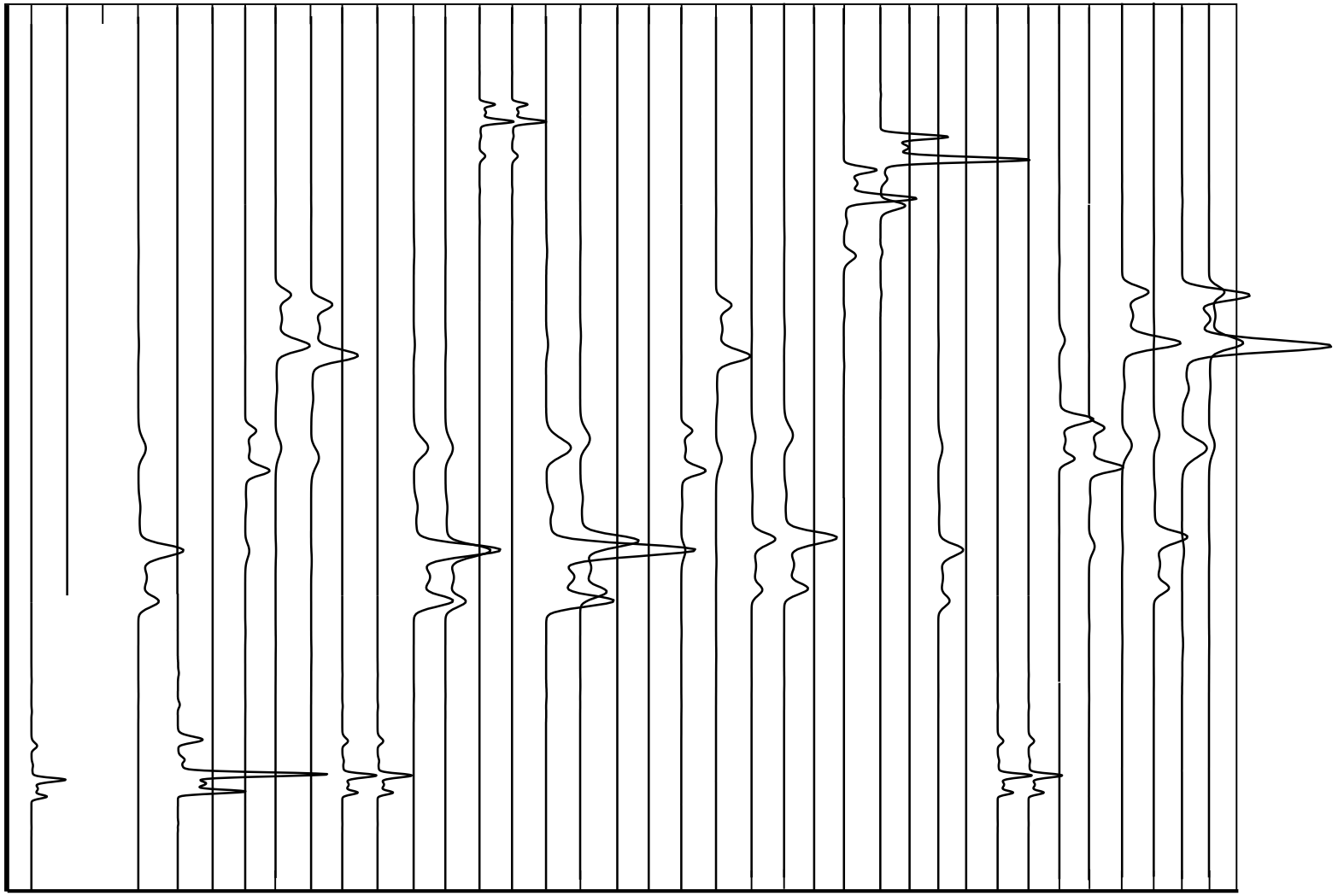


LECO's ChromaTOF Software

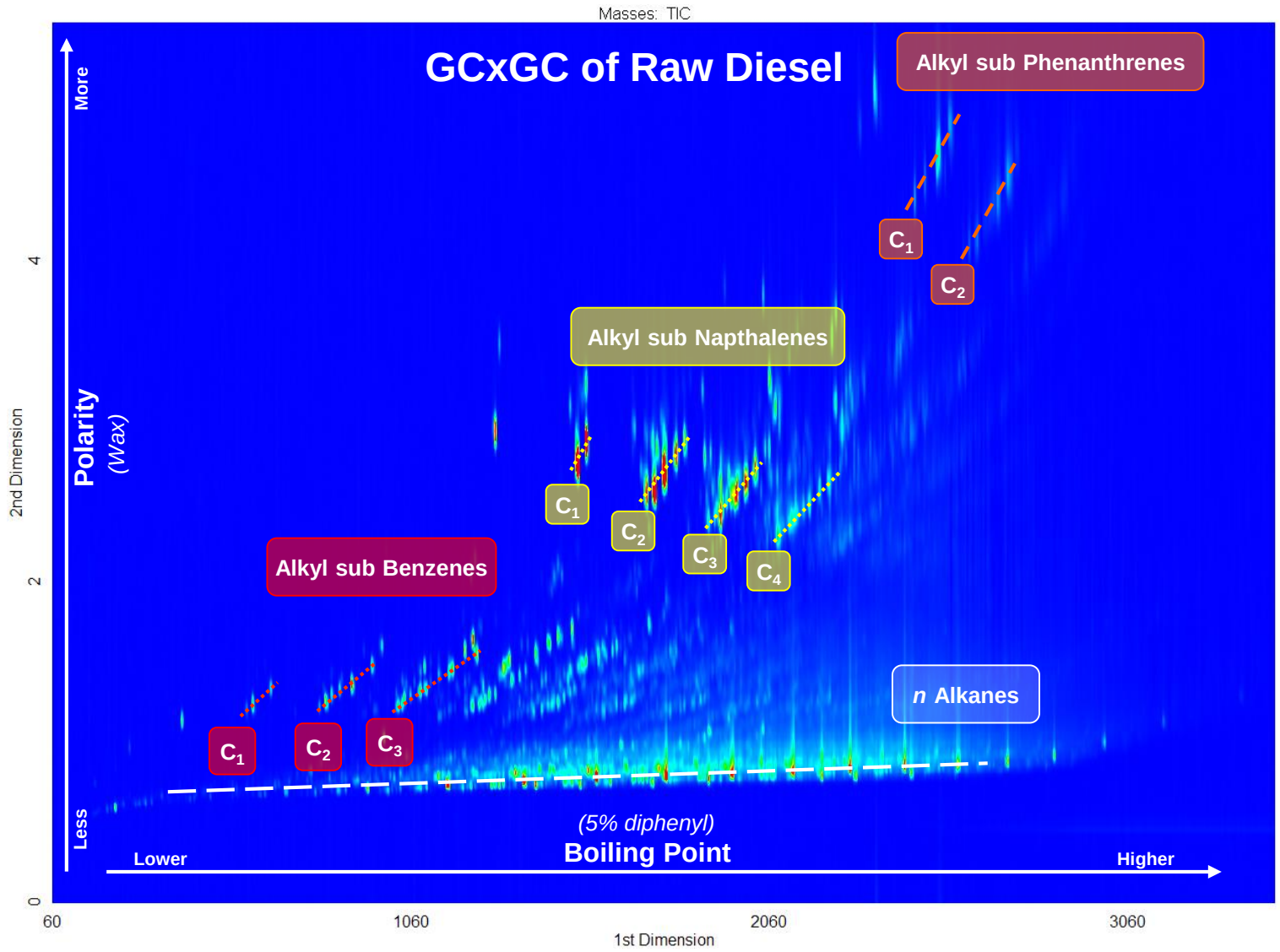
- True Signal Deconvolution
- Automated Peak Find
- Fully Integrated System Control
  - Acquisition method
  - Data processing
    - Qualitative processing
    - Quantative processing
- Statistical Compare Add-On Package
  - Peak alignment
  - Fisher ratios
- Easy Export of Data Files to Peripheral Software Packages



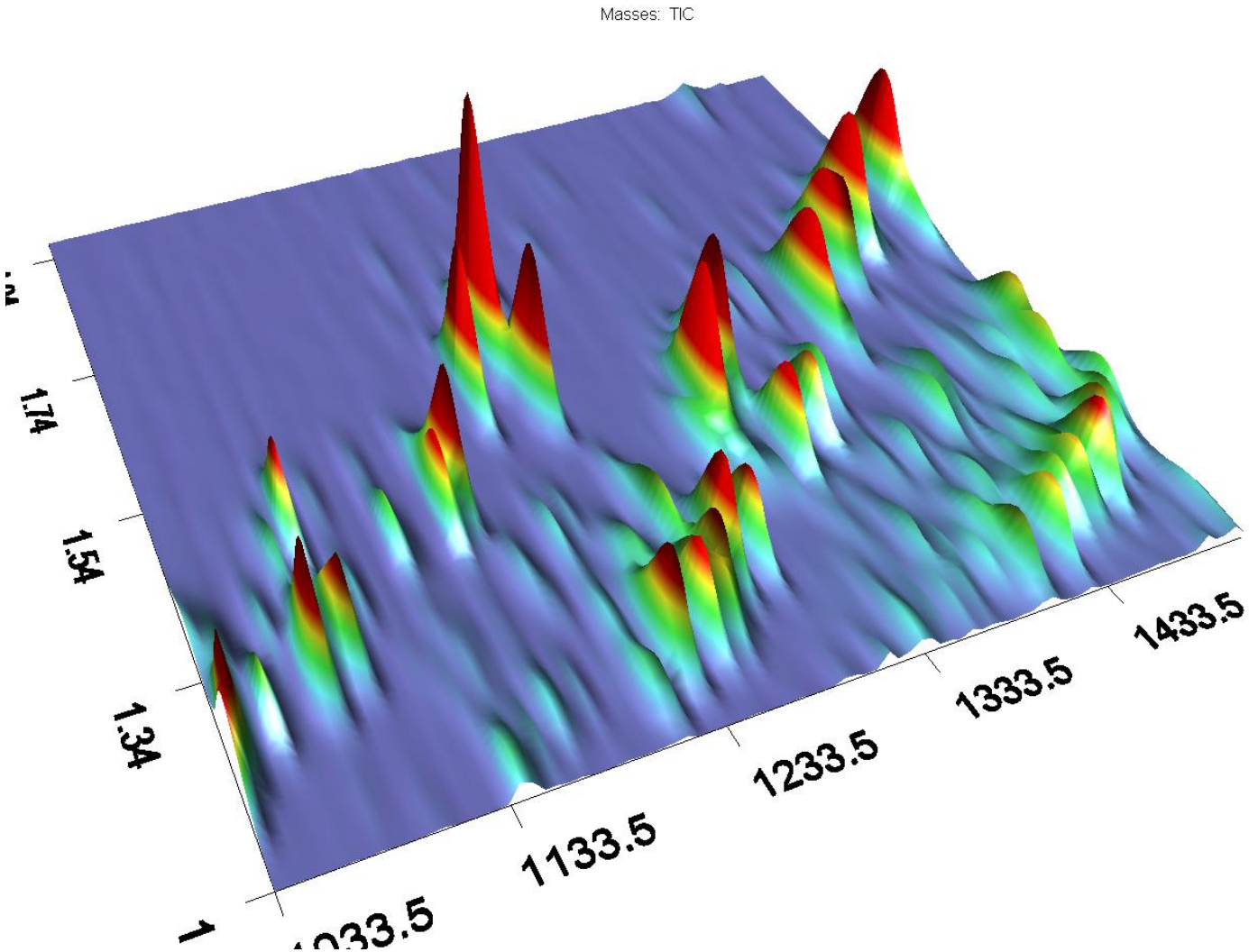
Retention Plane



Features of a GCxGC Contour Plot



Surface Plot



Study: ***SPE and GCxGC TOF-MS Analysis of Endocrine Disruptors and Other Pollutants from a Midwestern U.S. River System***

METHODS

- 13 solid phase extractions were completed on 1 liter aqueous samples
- Samples were obtained from 6 different rural and urban point sources along a Midwestern watershed
- Sample prep was via Solid Phase Extraction with an HLB hydrophilic modified styrene based polymer
  - *Selectivity advantage for a broad range of polar and nonpolar compounds from aqueous samples*
- Used EPA method reference standards:
  - *108 component reference standard used*

SPE Standard Method Development

- EDC Standard: EPA method 527 pesticide mix #1, method 551.1 pesticide/herbicide mix, method 8270 megamix, (Restek Corp.) and Bisphenol A (Sigma Aldrich)

  (108 compounds)

- SPE extraction method developed with 0.5 liter water spiked with 50ppb of EDC standard.

Solid Phase Extraction Procedure

- Adjust 1 liter water sample to pH 2 with 37% HCl.
- Condition SPE Supel™ Select HLB (Supelco Analytical, Sigma-Aldrich), 500mg cartridge with 5mL HPLC Water/5%Methanol, then 5mL Acetone, followed by 5mL HPLC water.
- Load 1 Liter of water using the Supelco Visiprep Vacuum Manifold (Supelco Analytical, Sigma–Aldrich) slowly unto the SPE cartridge.
- Dry SPE tube with vacuum for approximately 15 minutes.
- Elute slowly, 3mL of acetone/5% methanol into a 20mL clean glass test tube.
- Elute slowly, 3mL of dichloromethane into the same 20mL glass test tube.
- Speedvac to dryness for approximately 2 hours.
- Reconstitute dried residue in 500µL of acetone, vortex, and pipet into autosampler vial.
- Inject 1uL sample for GCxGC-TOFMS analysis.

GCxGC – TOFMS method development

GCxGC Analysis Parameters

- Gas Chromatograph: Agilent 7890 equipped with a LECO dual stage, quad jet thermal modulator and a GERSTEL MPS2 autosampler
- GC Primary Column: 30 m x 0.25 mm id. x 0.25 µm film thickness Rxi-5Sil-MS (Restek Corp.)
- GC Secondary Column: 1.0m x 0.18 mm id. X 0.18 µm film thickness Rxi-17Sil-MS (Restek Corp.)
- Carrier gas: Helium set @ 1.5 mL/min
- Injection mode: Splitless
- Injection volume: 1 µL
- Inlet temperature: 250°C
- Primary column temperature program: Initial temperature 75ºC for 1.0min. ramped @ 6.0°C/min. to 300ºC held for 10 min.
- Secondary column temperature program: Initial temperature 80ºC for 1.0min. ramped @ 6.0°C/min. to 305ºC held for 10 min.
- GCxGC modulator temperature offset: 20ºC
- Transfer Line Temperature: 280ºC
- Total run time: 48.50 min.

TOFMS Method

- Mass range: 35 – 800 u
- Acquisition rate: 200 spectra/s
- Ion source temperature: 230 °C
- Detector voltage: 1700 V
- Acquisition delay: 150 sec.

GCxGC Variable Modulation

Variable modulation was utilized shown which allows the user to change and modify the modulation period, hot pulse, and cool time throughout the GCxGC method to optimize the separation thereby maximizing the peak capacity in the chromatographic plane.

Modulation Timing: For 1D GC set second dimension time to 0

#	Start	End	Modulation Period (s)	Hot Pulse Time	Cool Time Between Stages
1st	Start of Run	614 s	2.00	0.50	0.50
2	614 s	1838 s	3.00	0.60	0.90
3	1838 s	End of Run	4.00	0.70	1.30

EDC Analysis Method

1L H20 pH 2 +
50ppb EDC Std
(108 compounds)
or water sample

→ Conditioned
500 mg SPE HLB

→ Dry

→ Elute
Acetone/
Methanol

→ Elute
Dichloromethane

↓

Speedvac-dry

↓

Reconstitute
500uL acetone

← 1uL
(5ng)

Pegasus® 4D (GCxGC-TOFMS)

Reference: Horizon Technology automated sample preparation
SPE Disk Cross Reference Guide

EDC Study Parameters

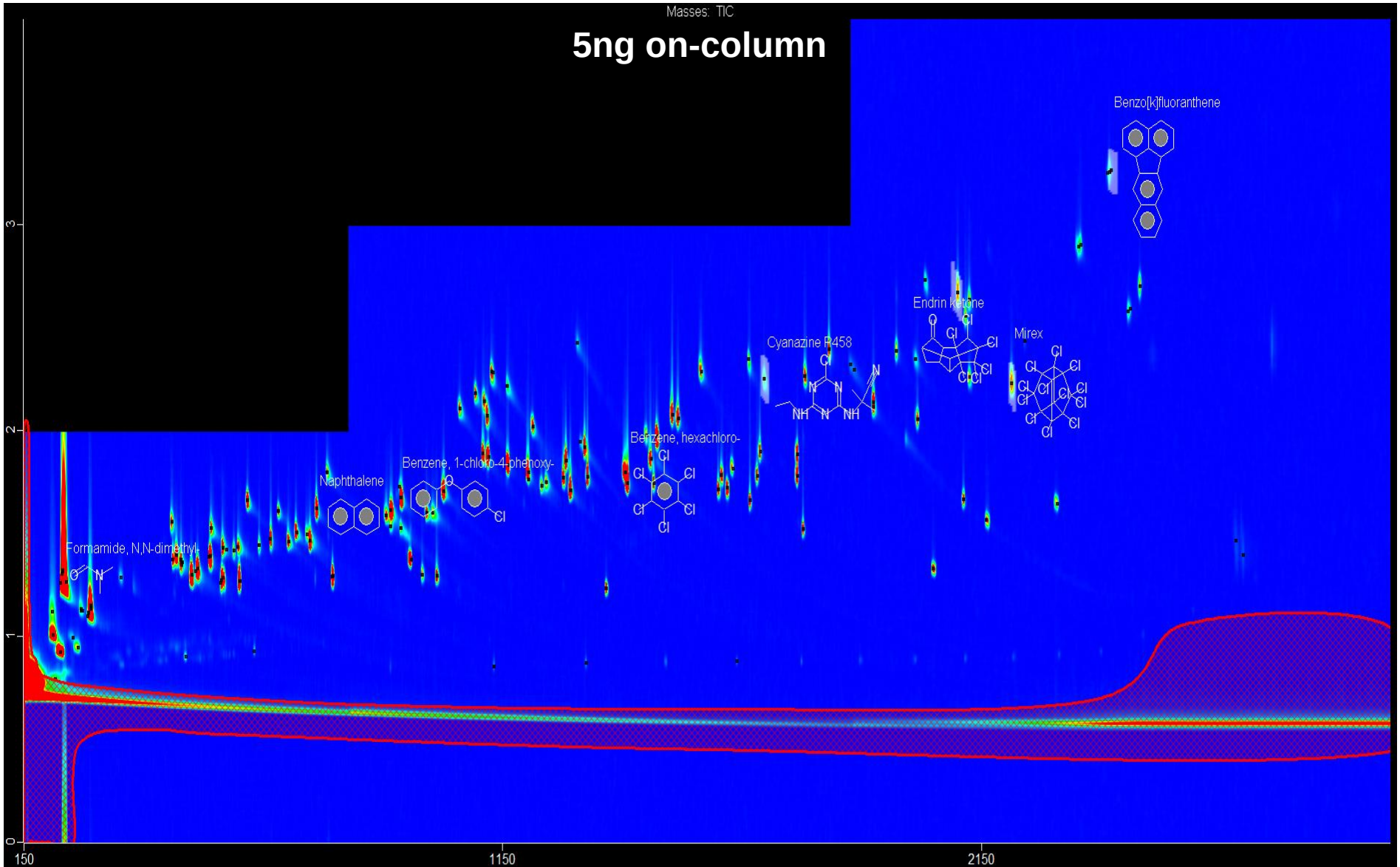
- 6 - Rural and urban locations along the watershed were sampled from Lake Michigan to approximately 50 miles upstream.
- 13 - 1 Liter samples were obtained
- GCxGC-TOFMS was carried with 2 injections per sample for a total of 26 analyses.

Data Analysis

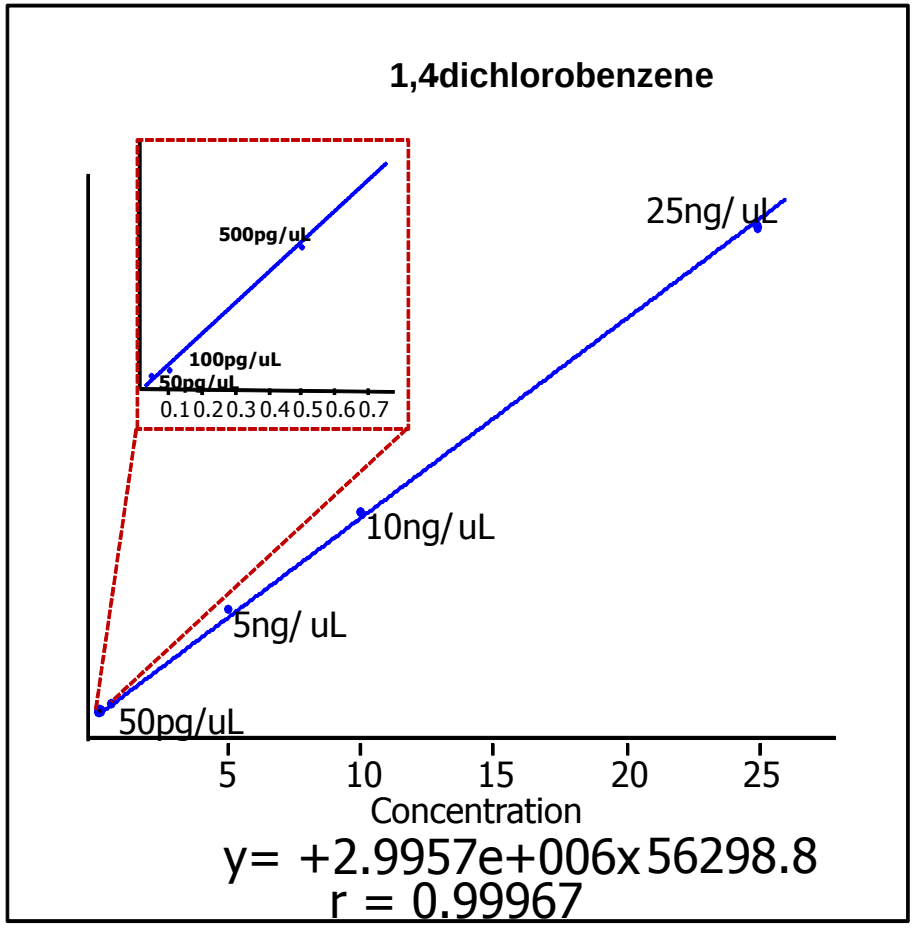
- MS library search (NIST, WILEY)
- Targeted quantification for selected analytes using calibration curves developed from the EDC standard
- Additionally, looked for untargeted analytes in the watershed samples

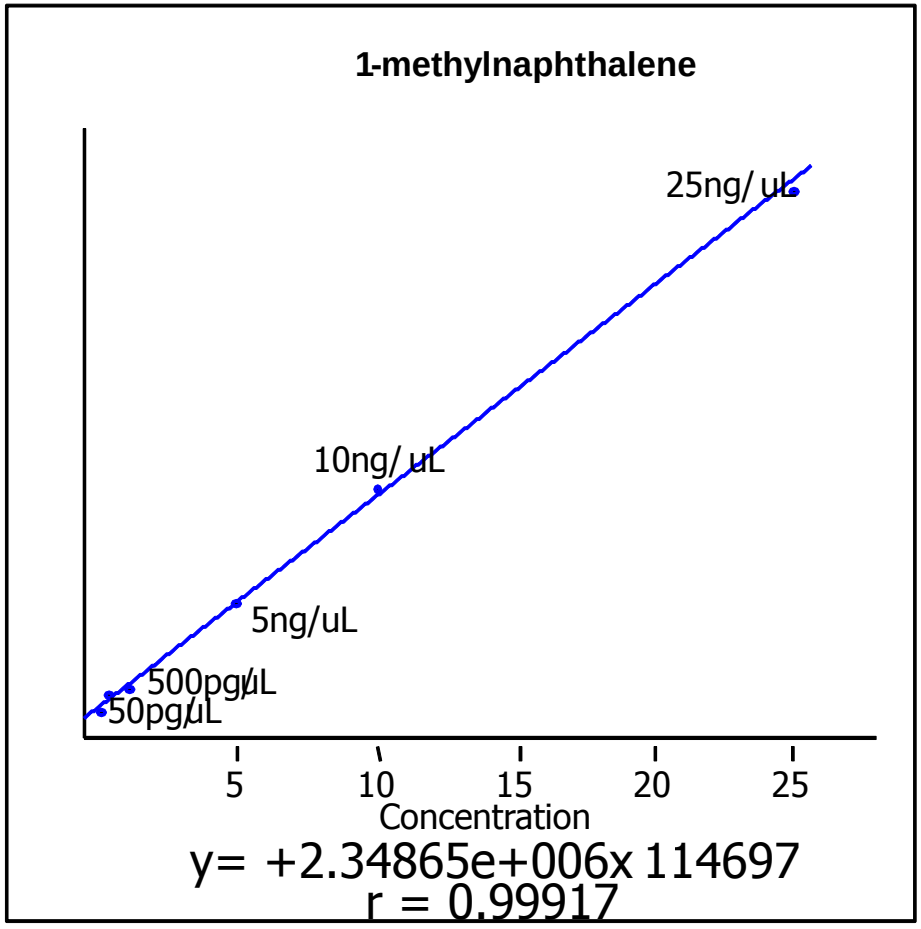
RESULTS

EDC Reference Std GCxGC Chromatogram



Selected Quantitative Calibrations





Name	Concentration	Conc. Units	R.T. (s)	Similarity	Area	Height	UniqueMass	S/N	Library
1,4-Dichlorobenzene	0.08	ng	396 , 1.345	941	169194	12113	146	364.46	Wiley9
Naphthalene, 1-methyl- (CAS)	0.06	ng	791 , 1.830	732	7533.1	605.24	141	70.566	Wiley9

Using the ChromaTOF Reference Feature

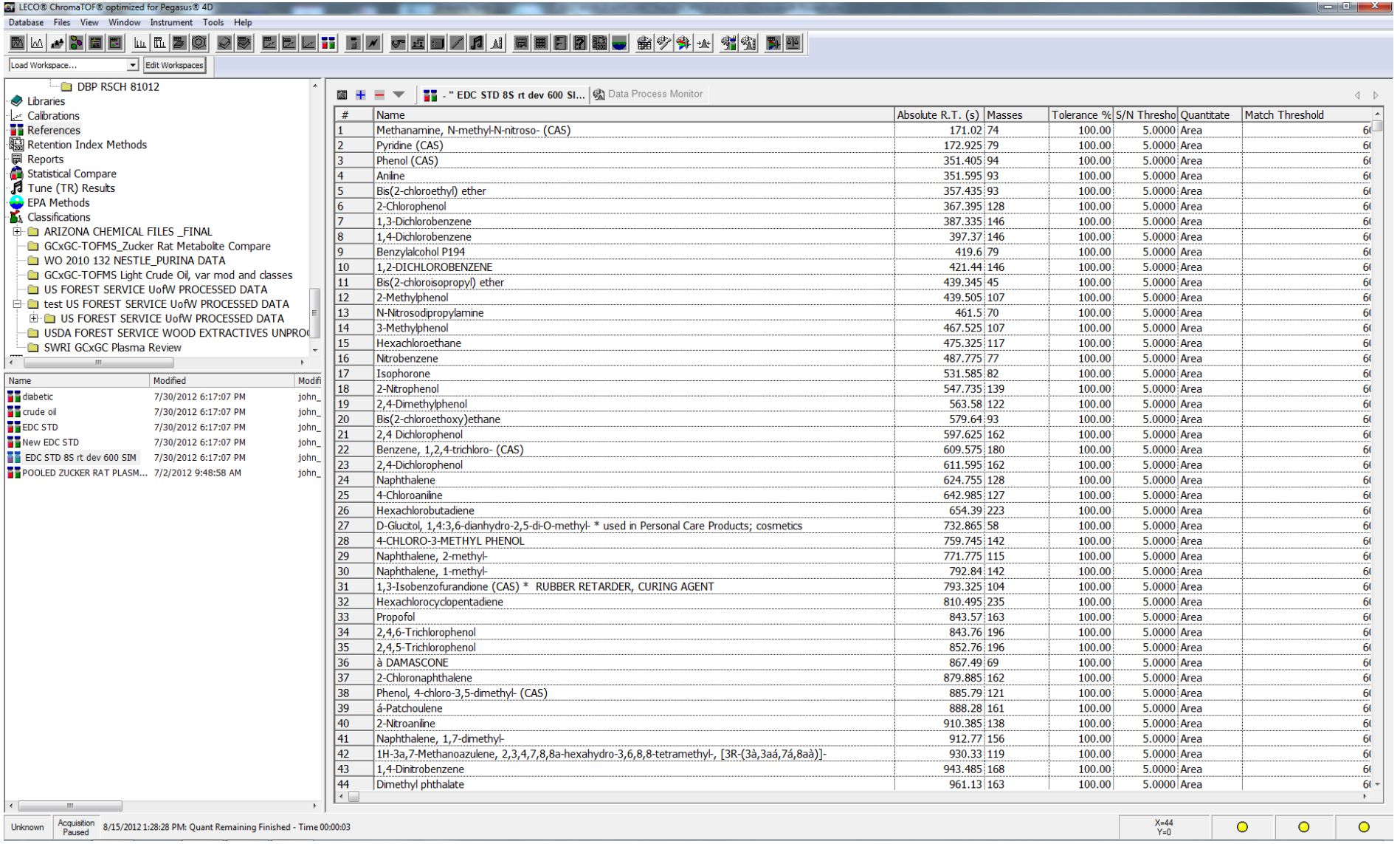
- A Reference Method is built in ChromaTOF® software from a user created standard that is applied and compared to a sample
- The purpose of a Reference Method is to determine the component differences between a sample and a reference standard within user defined limits of retention time, peak area, and spectral match.
- 108 component EDC standard used.
- Library identified components were ADDED to the Reference from analyzed samples as they were found.
- Final Reference used for data analysis contained 152 compounds.
- “Reference” is applied as part of the data processing method.

“ChromaTOF” REFERENCE FEATURE

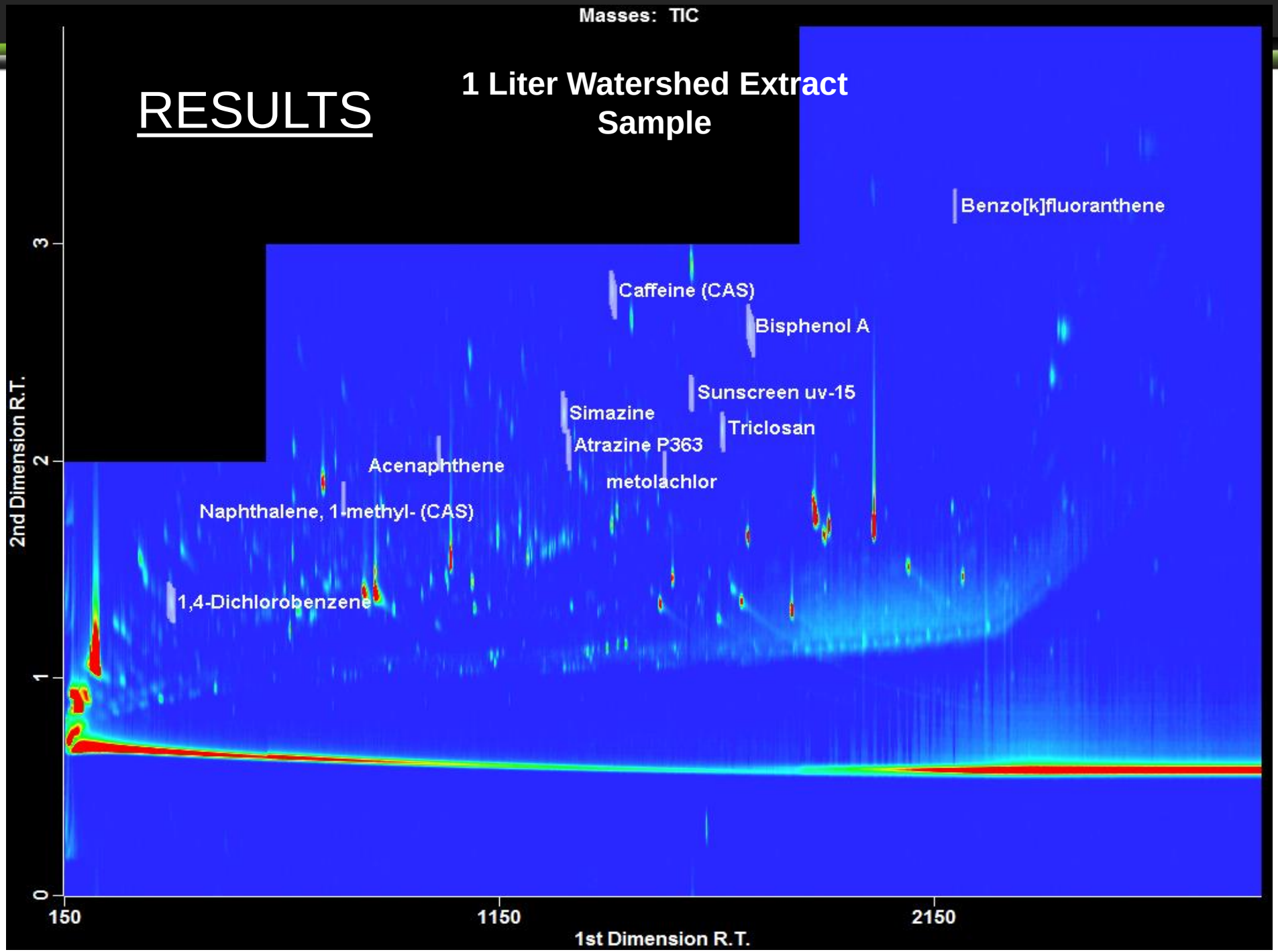
- The processed sample peak table displays each compound from the reference in the peak table. In the Type column the component is labeled as either a “Match”, found but “Out of Tolerance” by area percent, or “Not Found”.

Name	Type	Match	R.T. (s)	Area	UniqueMass	S/N	Library
Bisphenol A	Match	895	1724 , 2.600	740588	213	2979.2	mainlib
Ionol 2 * (ANTIOXIDANT- TO PREVENT GUMMING IN FUELS)	Out of Tolerance	918	1085 , 1.450	733755	219	5824.1	mainlib
Lindane/ EDC STD 8S rt dev 600 SIM	Not Found	600					mainlib

ChromaTOF EDC “Reference”



#	Name	Absolute R.T. (s)	Masses	Tolerance %	S/N Thresho	Quantitate	Match Threshold
1	Methanamine, N-methyl-N-nitroso- (CAS)	171.02	74	100.00	5.0000	Area	6
2	Pyridine (CAS)	172.925	79	100.00	5.0000	Area	6
3	Phenol (CAS)	351.405	94	100.00	5.0000	Area	6
4	Aniline	351.595	93	100.00	5.0000	Area	6
5	Bis(2-chloroethyl) ether	357.435	93	100.00	5.0000	Area	6
6	2-Chlorophenol	367.395	128	100.00	5.0000	Area	6
7	1,3-Dichlorobenzene	387.335	146	100.00	5.0000	Area	6
8	1,4-Dichlorobenzene	397.37	146	100.00	5.0000	Area	6
9	Benzylalcohol P194	419.6	79	100.00	5.0000	Area	6
10	1,2-DICHLOROBENZENE	421.44	146	100.00	5.0000	Area	6
11	Bis(2-chloroisopropyl) ether	439.345	45	100.00	5.0000	Area	6
12	2-Methylphenol	439.505	107	100.00	5.0000	Area	6
13	N-Nitrosodipropylamine	461.5	70	100.00	5.0000	Area	6
14	3-Methylphenol	467.525	107	100.00	5.0000	Area	6
15	Hexachloroethane	475.325	117	100.00	5.0000	Area	6
16	Nitrobenzene	487.775	77	100.00	5.0000	Area	6
17	Isophorone	531.585	82	100.00	5.0000	Area	6
18	2-Nitrophenol	547.735	139	100.00	5.0000	Area	6
19	2,4-Dimethylphenol	563.58	122	100.00	5.0000	Area	6
20	Bis(2-chloroethoxy)ethane	579.64	93	100.00	5.0000	Area	6
21	2,4 Dichlorophenol	597.625	162	100.00	5.0000	Area	6
22	Benzene, 1,2,4-trichloro- (CAS)	609.575	180	100.00	5.0000	Area	6
23	2,4-Dichlorophenol	611.595	162	100.00	5.0000	Area	6
24	Naphthalene	624.755	128	100.00	5.0000	Area	6
25	4-Chloroaniline	642.985	127	100.00	5.0000	Area	6
26	Hexachlorobutadiene	654.39	223	100.00	5.0000	Area	6
27	D-Glucitol, 1,4:3,6-dianhydro-2,5-di-O-methyl- * used in Personal Care Products; cosmetics	732.865	58	100.00	5.0000	Area	6
28	4-CHLORO-3-METHYL PHENOL	759.745	142	100.00	5.0000	Area	6
29	Naphthalene, 2-methyl-	771.775	115	100.00	5.0000	Area	6
30	Naphthalene, 1-methyl-	792.84	142	100.00	5.0000	Area	6
31	1,3-Isobenzofurandione (CAS) * RUBBER RETARDER, CURING AGENT	793.325	104	100.00	5.0000	Area	6
32	Hexachlorocyclopentadiene	810.495	235	100.00	5.0000	Area	6
33	Propofol	843.57	163	100.00	5.0000	Area	6
34	2,4,6-Trichlorophenol	843.76	196	100.00	5.0000	Area	6
35	2,4,5-Trichlorophenol	852.76	196	100.00	5.0000	Area	6
36	à DAMASCONE	867.49	69	100.00	5.0000	Area	6
37	2-Chloronaphthalene	879.885	162	100.00	5.0000	Area	6
38	Phenol, 4-chloro-3,5-dimethyl- (CAS)	885.79	121	100.00	5.0000	Area	6
39	á-Patchoulene	888.28	161	100.00	5.0000	Area	6
40	2-Nitroaniline	910.385	138	100.00	5.0000	Area	6
41	Naphthalene, 1,7-dimethyl-	912.77	156	100.00	5.0000	Area	6
42	1H-3a,7-Methanoazulene, 2,3,4,7,8,8a-hexahydro-3,6,8,8-tetramethyl-, [3R-(3à,3aá,7á,8aà)]-	930.33	119	100.00	5.0000	Area	6
43	1,4-Dinitrobenzene	943.485	168	100.00	5.0000	Area	6
44	Dimethyl phthalate	961.13	163	100.00	5.0000	Area	6


Masses: TIC
RESULTS
1 Liter Watershed Extract Sample
Benzo[k]fluoranthene
Caffeine (CAS)
Bisphenol A
Sunscreen uv-15
Simazine
Triclosan
Atrazine P363
Acenaphthene
metolachlor
Naphthalene, 1-methyl- (CAS)
1,4-Dichlorobenzene
2nd Dimension R.T.
0
1
2
3
150
1150
2150
1st Dimension R.T.

EDC STUDY COMBINED RESULTS TABLE

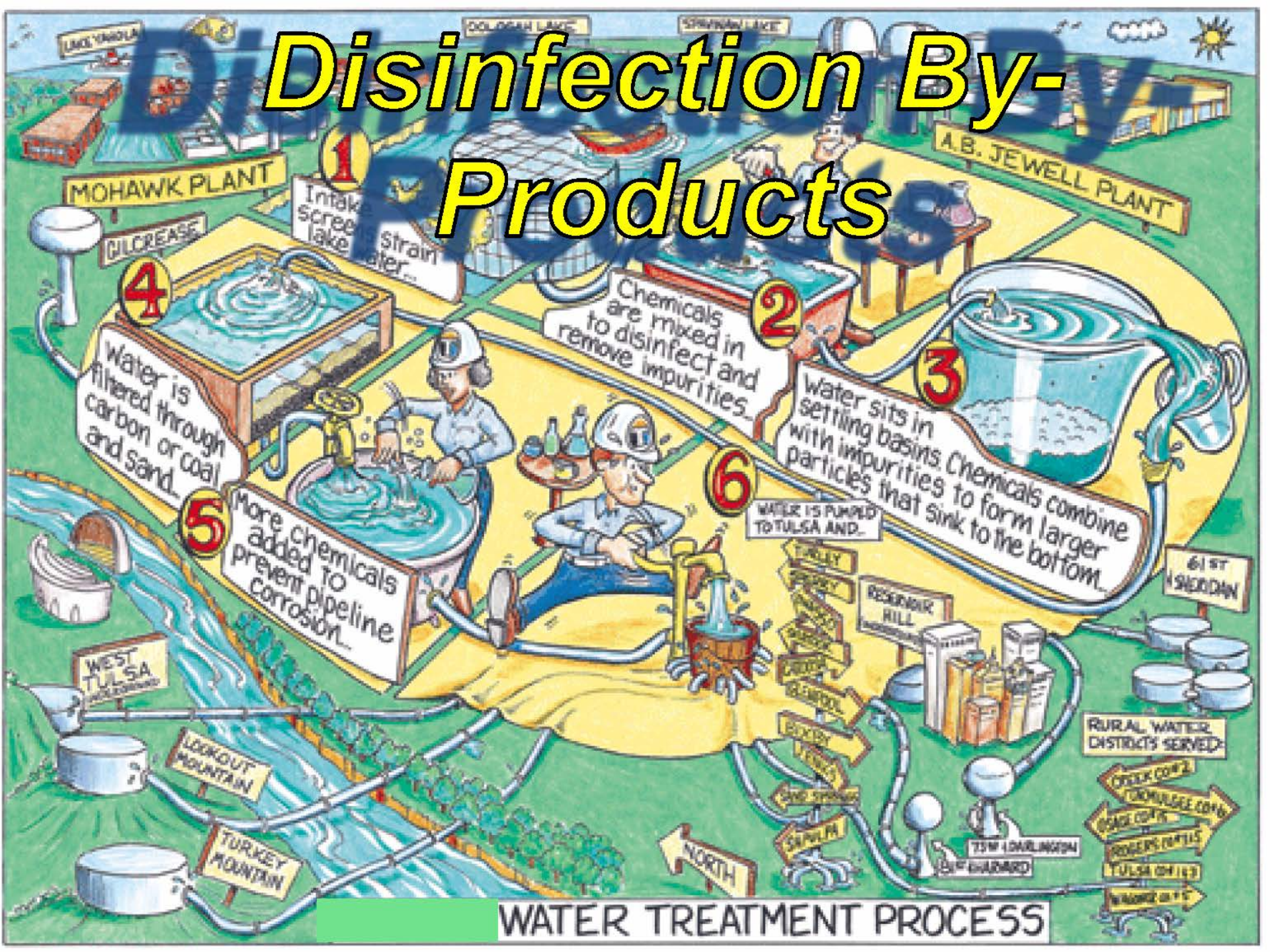
	Analytes Detected	1ST DIMENSION RETENTION TIME (s)	COUNT # OF TIMES DETECTED	CHEMICAL TYPE
1	Phenol (CAS)	354	23	EDC
2	Bis(2-chloroethyl) ether	364	1	EDC
3	1,4-Dichlorobenzene	392	21	EDC
4	1,3-Dichlorobenzene	394	9	EDC
5	1,2-DICHLOROBENZENE	418	3	EDC
6	Benzylalcohol P194	420	19	EDC
7	2-Methylphenol	440	22	EDC
8	3-Methylphenol	458	24	EDC
9	Nitrobenzene	480	7	EDC
10	Isophorone * Solvent	532	26	EDC, INDUSTRIAL CHEMICAL
11	2-Nitrophenol	548	11	EDC
12	2,4-Dimethylphenol	566	11	EDC
13	2,4 Dichlorophenol	600	7	EDC
14	Benzene, 1,2,4-trichloro- (CAS)	608	9	EDC
15	Naphthalene	623	25	EDC, PAH
16	D-Glucitol, 1,4:3,6-dianhydro-2,5-di-O-methyl- * used in Personal Care Products; cosmetics	731	7	PCP
17	Naphthalene, 2-methyl-	770	20	EDC, PAH
18	Naphthalene, 1-methyl-	791	22	EDC, PAH
19	1,3-Isobenzofurandione (CAS) * RUBBER RETARDER, CURING AGENT	794	14	EDC, INDUSTRIAL CHEMICAL
20	Propofol	842	6	PCP, PHARMACEUTICAL
21	2,4,6-Trichlorophenol	842	6	EDC
22	2,4,5-Trichlorophenol	845	6	EDC
23	à DAMASCONE	863	12	FOOD, FLAVOR, FRAGRANCE
24	2-Chloronaphthalene	878	7	EDC, PAH
25	Phenol, 4-chloro-3,5-dimethyl- (CAS)	884	6	EDC
26	á-Patchoulene	887	7	FOOD, FLAVOR, FRAGRANCE
27	Naphthalene, 1,7-dimethyl-	911	7	EDC, PAH
28	1H-3a,7-Methanoazulene, 2,3,4,7,8,8a-hexahydro-3,6,8,8-tetramethyl-, [3R-(3à,3aá,7á,8aà)]-	929	5	EDC
29	Acenaphthylene (CAS)	971	14	EDC, PAH
30	Acenaphthene	1010	21	EDC, PAH

31	BUTYL HYDROXY TOLUENE * BHT ANTIOXIDANT	1025	23	EDC, FOOD, FLAVOR, FRAGRANCE
32	Tributyl phosphate * SOLVENT & PLASTICIZER	1031	19	EDC, INDUSTRIAL CHEMICAL
33	Benzeneacetic acid, ethyl ester * flavor fragrance	1037	25	EDC, FOOD, FLAVOR, FRAGRANCE
34	à-N-METHYL IONONE	1043	6	EDC, FOOD, FLAVOR, FRAGRANCE
35	TRIPROPYLENE GLYCOL 5	1046	21	EDC, INDUSTRIAL CHEMICAL
36	Dibenzofuran	1052	14	EDC
37	Lilial	1055	12	PCP, FOOD, FLAVOR, FRAGRANCE
38	1H-Benzotriazole, 4-methyl- * CORROSION INHIBITOR	1070	4	EDC, INDUSTRIAL CHEMICAL
39	1H-Benzotriazole, 5-methyl- *RETROCURE G USED IN PREVULCANIZATION IN RUBBER MANUFATURE	1076	4	EDC, INDUSTRIAL CHEMICAL
40	Ionol 2 * (ANTIOXIDANT- TO PREVENT GUMMING IN FUELS)	1085	11	EDC, INDUSTRIAL CHEMICAL
41	2-tert-Butylhydroquinone * TBHQ, FOOD PRESERVATIVE ANTIOXIDANT	1088	2	FOOD, FLAVOR, FRAGRANCE
42	Dodecanamide, N,N-bis(2-hydroxyethyl)- •FOAM STABILIZER IN HOUSEHOLD DETERGENTS AND SHAMPOOS	1091	22	PCP
43	á N METHYL IONONE	1109	6	PCP, FOOD, FLAVOR, FRAGRANCE
44	DEET * INSECTICIDE	1112	26	PCP, EDC, INSECTICIDE
45	Gabapentin	1127	13	PCP, PHARMACEUTICAL
46	Fluorene	1130	16	EDC, PAH
47	4-Chlorophenyl phenyl ether	1139	5	EDC, FLAME RETARDANT
48	Benzothiazole, 2-(methylthio)- (CAS)	1151	14	EDC
49	Ibuprofen	1160	5	PCP, PHARMACEUTICAL
50	2,6-Bis(1,1-dimethylethyl)-4-(1-oxopropyl)phenol	1166	23	EDC, UV LIGHT STABILIZER
51	Diphenylamine * ANTIOXIDANT, SCALD INHIBITIOR USED ON APPLES	1169	2	EDC, FOOD, FLAVOR, FRAGRANCE
52	Azobenzene	1175	9	EDC
53	Kayacure bp * used in the manufacturing of antihistamines, hypnotics, insecticides.	1178	11	EDC, PHARMACEUTICAL, INSECTICIDE
54	N,N,N',N'-Tetraacetylethylenediamine *LIGAND FOR METAL IONS, ACRYLAMIDE POLYMERIZATION	1178	10	EDC, INDUSTRIAL CHEMICAL
55	Clovene	1181	18	FOOD, FLAVOR, FRAGRANCE
56	TRANS-METHYL DIHYDROJASMONATE	1196	21	EDC, FOOD, FLAVOR, FRAGRANCE
57	Trifluralin	1214	6	EDC, HERBICIDE
58	4-Bromophenyl phenyl ether *USED AS A (PAST) FLAME RETARDANT	1250	4	EDC, BFR, INDUSTRIAL CHEMICAL
59	Ibuprofen-M (HO-) -H2O P329	1253	6	PCP, PHARMACEUTICAL
60	Hexachlorobenzene	1259	18	EDC, FUNGICIDE, POP, banned globally
61	Simazine	1298	14	EDC, HERBICIDE, banned by EU
62	Atrazine P363	1307	22	EDC, HERBICIDE, banned EU, US still uses
63	Myristic acid P412	1313	24	FOOD, FLAVOR, FRAGRANCE
64	Cedryl acetate	1331	10	PCP, FOOD, FLAVOR, FRAGRANCE

65	Ibuprofen-M (HO-) isomer-1 ME P444 * Pharmacuetical	1334	6	PCP, PHARMACEUTICAL
66	Tris(1-chloro-2-propyl)phosphate *TCPP Flame retardant	1349	26	EDC, INDUSTRIAL CHEMICAL
67	Phenanthrene	1352	24	EDC, PAH
68	Anthracene	1358	11	EDC, PAH
69	Naphthalene, 6,7-diethyl-1,2,3,4-tetrahydro-1,1,4,4-tetramethyl- (CAS)	1364	13	EDC
70	3,5-DITERT-BUTYLBENZALDEHYDE	1373	7	EDC
71	Caffeine (CAS)	1406	10	PCP, FOOD, FLAVOR, PHARMA
72	Carbazole	1415	12	EDC, INDUSTRIAL CHEMICAL
73	7-Acetyl-6-ethyl-1,1,4,4-tetramethyltetralin * synthetic musk	1415	24	PCP, FOOD, FLAVOR, FRAGRANCE
74	Gemfibrozil * Medication to lower lipid levels	1490	11	PCP, PHARMACEUTICAL
75	metolachlor	1529	24	EDC, HERBICIDE
76	5H-dibenz[b,f]azepine (CAS) * Pharmaceutical - intermediate used in drug manufacturing	1550	9	PCP, PHARMACEUTICAL
77	Benzenesulfonanilide * production of synthetic dyes, photochemicals, disinfectants, electroplating	1589	8	EDC, INDUSTRIAL CHEMICAL
78	Sunscreen uv-15	1592	6	PCP, PHARMACEUTICAL
79	Heptachlor epoxide	1612	2	EDC, POP, INSECTICIDE, US limits use
80	Oxychlordane	1613	2	EDC, POP, INSECTICIDE, banned US 1988
81	Bioallethrin	1625	4	EDC, PESTICIDE
82	FLUORANTHENE	1628	23	EDC, PAH
83	Lauryl acrylate * for manufacturing polymers used in hairstyling	1640	6	PCP
84	Naproxen * Pharmaceutical	1655	1	PCP, PHARMACEUTICAL
85	Triclosan * antibacterial, antifungal agent used in toothpaste	1661	7	PCP, PHARMACEUTICAL
86	Pyrene	1679	23	EDC, PAH
87	Butyl citrate * used as a plasticizer, antifoam agent	1721	5	EDC, INDUSTRIAL CHEMICAL
88	Bisphenol A * Used in making plastics	1721	13	EDC, INDUSTRIAL CHEMICAL
89	Nitrofen	1769	2	EDC, PESTICIDE, banned in EU and US
90	Endrin P1112	1772	2	EDC, INSECTICIDE, banned 2004
91	TCPP _ Tris(1,3-dichloroisopropyl)phosphate * (PBDE) flame retardant REPLACEMENT	1838	16	EDC, INDUSTRIAL CHEMICAL
92	2H-1-Benzopyran-2-one, 7-(diethylamino)-4-methyl- * (Optical bleach in textile industry)	1854	4	EDC, INDUSTRIAL CHEMICAL
93	Hexazinone P513	1882	1	EDC, HERBICIDE
94	Bifenthrin	1958	5	EDC, INSECTICIDE, class C carcinogen
95	Benzo[a]anthracene	1962	4	EDC, PAH
96	Chrysene (CAS)	1970	8	EDC, PAH
97	Methoxychlor	1974	2	EDC, INSECTICIDE U.S. BANNED 2003
98	Mirex	2062	2	EDC, INSECTICIDE U.S. BANNED 1976
99	Benzo[b or k]fluoranthene	2198	15	EDC, PAH
100	Benzo[a]pyrene (CAS)	2262	3	EDC, PAH
101	Fenvalerate isomer-1 P1241	2302	1	EDC, INSECTICIDE
102	Fenvalerate isomer-2 P1242	2322	2	EDC, INSECTICIDE

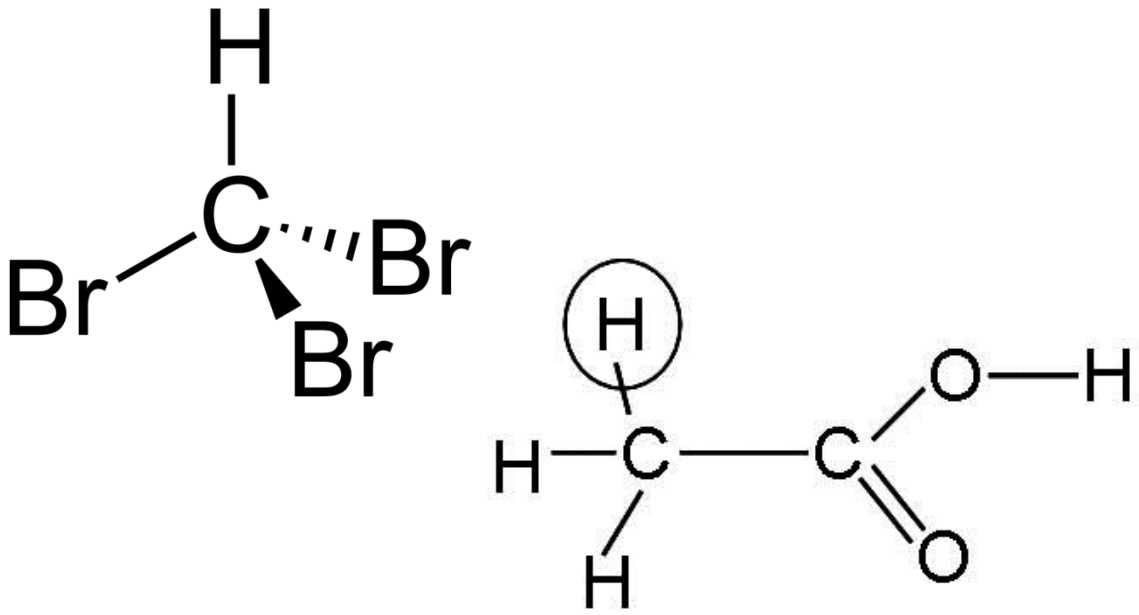
Conclusions to Watershed Study

- A total of 102 chemicals were detected from 6 point sources in a Midwestern watershed
- The compounds detected matched the reference standard with at least a 60% library match similarity
- Out of the 26 samples studied (13 samples x 2 replicates each), 81% of the 102 chemicals were found at least 5 times.


Disinfection By-Products
MOHAWK PLANT
A.B. JEWELL PLANT
1
Intake screen lake
Strain water
2
Chemicals are mixed in to disinfect and remove impurities.
3
Water sits in settling basins. Chemicals combine with impurities to form larger particles that sink to the bottom.
4
Water is filtered through carbon or coal and sand.
5
More chemicals added to prevent pipeline corrosion.
6
WATER IS PUMPED TO TULSA AND...
TURLEY
PERKY
RESERVOIR HILL
61ST SHERIDAN
WEST TULSA
LOOKOUT MOUNTAIN
TURKEY MOUNTAIN
NORTH
SAPULPA
RURAL WATER DISTRICTS SERVED
WATER TREATMENT PROCESS

DBPs— Formation

• Disinfection byproducts can form when disinfectants, such as chlorine, react with naturally present compounds in the water.



Read more:
http://www.lenntech.com/processes/disinfection/byproducts/disinfection-byproducts.htm#ixzz23oaB12iE

Drinking Water DBPs— What are the issues?

- Concern over possible human health risk:

- Epidemiologic studies: risk of bladder cancer; some cause cancer in laboratory animals

- Recent concerns about possible reproductive & developmental effects (from epidemiology studies)

- Goal of EPA research: Comprehensively identify DBPs formed from different disinfectants, test for toxicity, understand their formation, minimize or eliminate in drinking water

DBP Health Risk???

"Some have suggested an increased cancer risk to those exposed to chlorinated waters while others have demonstrated none. In issuing the Stage 1 Disinfectants/Disinfection Byproducts Rule based on evidence then available, EPA stated that while the agency cannot conclude there is a causal link between exposure to chlorinated surface water and cancer, these studies have suggested an association, albeit small, between bladder, rectal, and colon cancer and exposure to chlorinated surface water. "

Table 1

MRDLGs, MRDLs, MCLGs and MCLs for Stage 1 Disinfectants and Disinfection Byproducts Rule

DISINFECTANT RESIDUAL	MRDLG (mg/L)	MRDL (mg/L)	COMPLIANCE BASED ON
Chlorine	**4 (as Cl_2)**	**4.0 (as Cl_2)**	**Annual Average**
Chloramine	**4 (as Cl_2)**	**4.0 (as Cl_2)**	**Annual Average**
Chlorine Dioxide	**0.8 (as ClO_2)**	**0.8 (as ClO_2)**	**Daily Samples**
DISINFECTION BYPRODUCTS	MCLG (mg/L)	MCL (mg/L)	COMPLIANCE BASED ON
Total trihalomethanes (TTHM)[1]	**N/A**	**0.080**	**Annual Average**
– Chloroform	***		
– Bromodichloromethane	**0**		
– Dibromochloromethane	**0.06**		
– Bromoform	**0**		
Haloacetic acids (five) (HAA5)[2]	**N/A**	**0.060**	**Annual Average**
– Dichloroacetic acid			
– Trichloroacetic acid	**0**		
	0.3		
Chlorite	**0.8**	**1.0**	**Monthly Average**
Bromate	**0**	**0.010**	**Annual Average**

References - http://water.epa.gov/lawsregs/rulesregs/sdwa/stage1/factsheet.cfm
http://www.epa.gov/enviro/html/icr/dbp_health.html

Samples

- Potable water
- Residential water: 4 samples, 8 analyses
- Lab tap water: 2 samples, 4 analyses
- Bottled Water: 1 sample, 2 analyses
- Swimming pool water
- Residence #1: 2 samples, 4 analyses
- Residence #2: 2 samples, 4 analyses

METHODS

- SAMPLE PREPARATION: 1 Liter extraction using SPE HLB washed resin in glass cartridges with TEFLON frits.

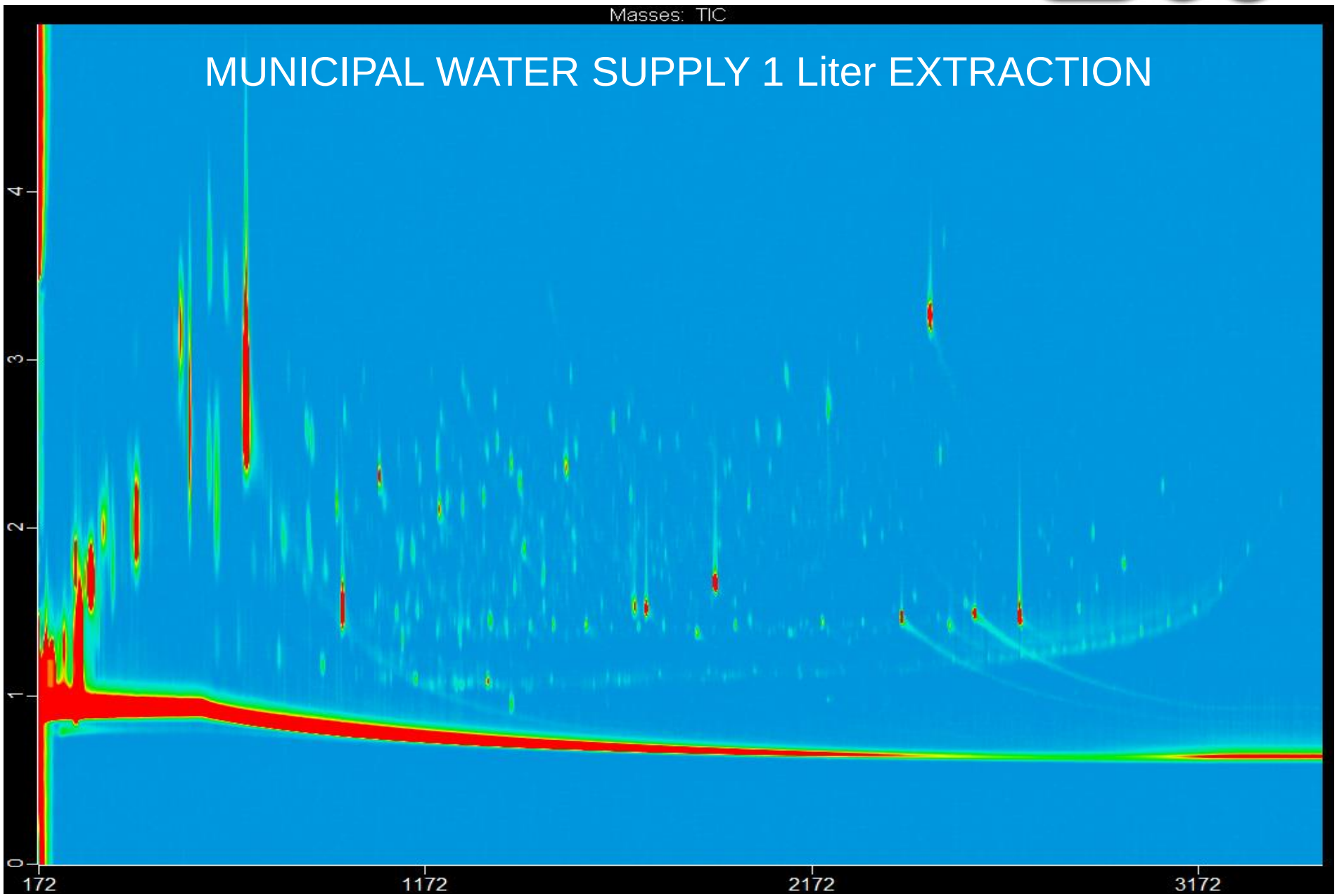
- GCxGC-TOFMS analysis:

GCxGC Analysis Parameters

- Gas Chromatograph: Agilent 7890 equipped with a LECO dual stage, quad jet thermal modulator and a GERSTEL MPS2 autosampler
- GC Primary Column: 30 m x 0.25 mm id. x 0.25 µm film thickness Rxi-5Sil-MS (Restek Corp.)
- GC Secondary Column: 1.25m x 0.18 mm id. X 0.18 µm film thickness Rxi-17Sil-MS (Restek Corp.)
- Carrier gas: Helium set @ 1.0 mL/min
- Injection mode: Splitless
- Injection volume: 1 µL
- Inlet temperature: 250°C
- Primary column temperature program: Initial temperature 30ºC for 10.0min. ramped @ 6.0°C/min. to 295ºC held for 4 min.
- Secondary column temperature program: Initial temperature 40ºC for 10.0min. ramped @ 6.0°C/min. to 305ºC held for 10 min.
- GCxGC modulator temperature offset: +15ºC
- Transfer Line Temperature: 250ºC
- Total run time: 58.17 min.

TOFMS Method

- Mass range: 35 – 800 u
- Acquisition rate: 200 spectra/s
- Ion source temperature: 230 °C
- Detector voltage: 1425 V
- Acquisition delay: 192 sec.


Masses: TIC
MUNICIPAL WATER SUPPLY 1 Liter EXTRACTION
4
3
2
1
0
172
1172
2172
3172

Municipal Water Supply ***RESULTS***
EPA Method 551.1 & Priority DBPs from National Occurrence Study List

Halomethanes

Chloroform
Methane, bromo-
Methane, bromodichloro-
Methane, dibromochloro-
Methane, tribromo-
Dichloroiodomethane

Haloacetonitriles

Acetonitrile, bromochloro-
Acetonitrile, chloro-
Acetonitrile, dibromo-
Acetonitrile, dichloro-

Haloketones

2-Propanone, 1,3-dichloro-
2-Propanone, 1,1,1-trichloro-
2-Propanone, 1-chloro-
1,1,1,3,3-Pentachloropropanone

Haloaldehydes

Dichloroacetaldehyde
Bromochloroacetaldehyde
Triclhoroacetaldehyde monohydrate (Chloral Hydrate)

Halonitromethanes

Dichloronitromethane
Bromochloronitromethane

Haloamides

Dibromoacetamide
Acetamide, 2,2-dichloro-
Acetamide, 2,2,2-trichloro-

Haloacetic acids

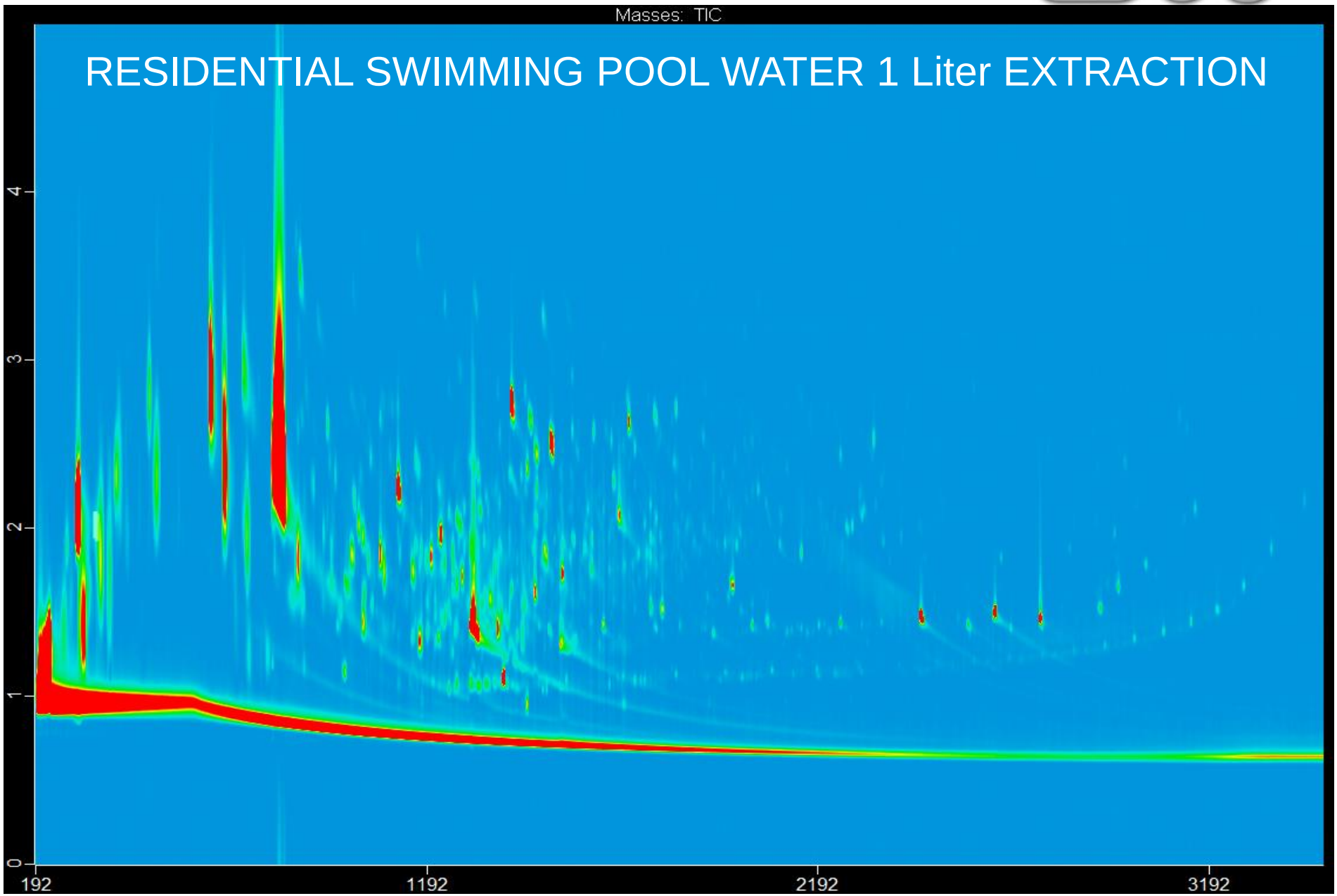
Acetic acid, dichloro-
Bromochloroacetic acid
Monochloroacetic acid
Trichloroacetic acid

Haloacids

3,3-Dichloropropenic acid

VOCs and Miscellaneous DBPs

Benzyl chloride
Trichloroethylene
Chlorodibromoacetaldehyde
Benzene, 1,4-dichloro-


Masses: TIC
RESIDENTIAL SWIMMING POOL WATER 1 Liter EXTRACTION
4
3
2
1
0
192
1192
2192
3192

Residential Swimming Pool Water ***RESULTS***

EPA Method 551.1 & Priority DBPs from National Occurrence Study List

Halomethanes

Chloroform
Methane, bromo-
Methane, bromodichloro-
Methane, dibromochloro-
Methane, tribromo-
~~Dichloroiodomethane~~

Haloacetonitriles

Acetonitrile, bromochloro-~~Acetonitrile, chloro-~~
Acetonitrile, dibromo-
Acetonitrile, dichloro-

Haloketones

2-Propanone, 1,3-dichloro-
2-Propanone, 1,1,1-trichloro-
2-Propanone, 1-chloro-
1,1,1,3,3-Pentachloropropanone
2-Propanone, 1,1,3,3-tetrachloro-
2-Propanone, 1,1,1,3-tetrachloro-
1-Bromo-1,1-dichloro-2-propanone

Haloaldehydes

Dichloroacetaldehyde
~~Bromochloroacetaldehyde~~
Trichloroacetaldehyde monohydrate (Chloral Hydrate)

Halonitromethanes

Dichloronitromethane
~~Bromochloronitromethane~~
Trichloronitromethane (Chloropicrin)

Haloamides

~~Dibromoacetamide~~
Acetamide, 2,2-dichloro-
Acetamide, 2,2,2-trichloro-

Haloacetic acids

Acetic acid, dichloro-
Bromochloroacetic acid
Monochloroacetic acid
Trichloroacetic acid
Acetic acid, bromo-

Haloacids

3,3-Dichloropropenic acid

VOCs and Miscellaneous DBPs

Benzyl chloride
Trichloroethylene
~~Chlorodibromoacetaldehyde~~
Benzene, 1,4-dichloro-
Phenol, 2,4-dichloro-
Phenol, 2,4,6-trichloro-

EDCs Found in Municipal Water, Swimming Pools, and Bottled Drinking Water Compared to the TEDX List of Potential Endocrine Disruptors

Name	R.T. (s)	Similarity	Area	UniqueMass	S/N	Library
Benzene (CAS)	262 , 1.435	845	4009119	78	6108.3	Wiley9
1,1'-Biphenyl (CAS)	1807 , 2.020	789	15184	154	287.41	Wiley9
Benzophenone	2112 , 2.365	818	104047	105	1251.3	replib
Phenol, p-tert-butyl-	1677 , 1.780	864	99384	135	1483.7	mainlib
Toluene	502 , 2.305	943	9473479	91	9374.3	replib
Bisphenol A	2657 , 2.950	612	19381	213	159.08	Wiley9
Naphthalene (CAS)	1527 , 2.015	702	17635	128	295.38	Wiley9
Phenol	1162 , 1.890	913	307236	94	1583.5	mainlib
Phenol, 4-(1,1,3,3-tetramethylbutyl)- (CAS)	2077 , 1.765	676	26230	135	517.82	Wiley9
p-Cresol	1342 , 1.845	718	18601	107	210.8	replib

LSCA Bottled Drinking Water ***RESULTS***
EPA Method 551.1 & Priority DBPs from National Occurrence Study List

Halomethanes

Chloroform
Methane, bromodichloro-
Methane, dibromochloro-
Methane, tribromo-

Haloaldehydes

Haloacetic acids

Haloacetonitriles

Halonitromethanes

Haloacids

Haloketones

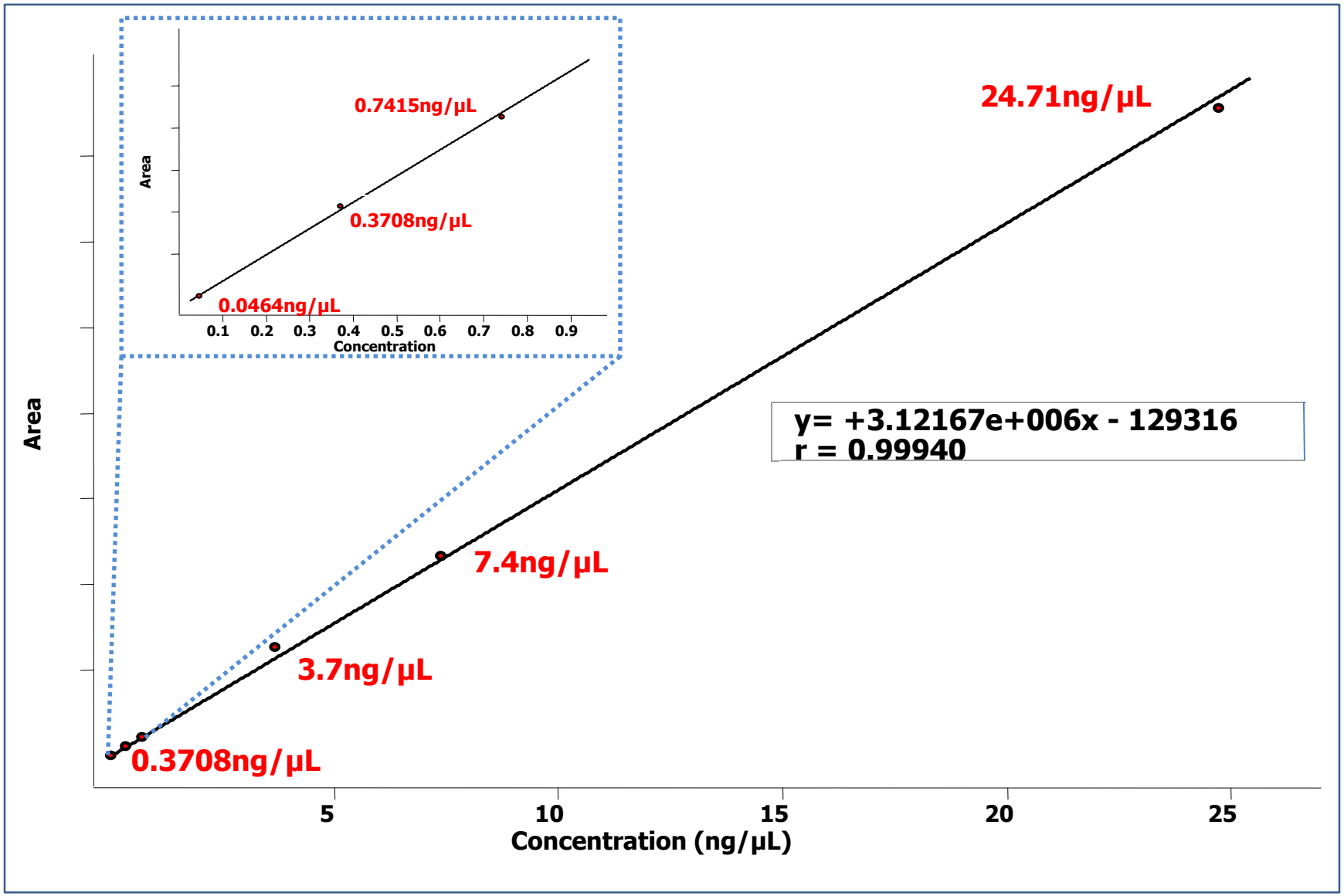
2-Propanone, 1-chloro-

Haloamides

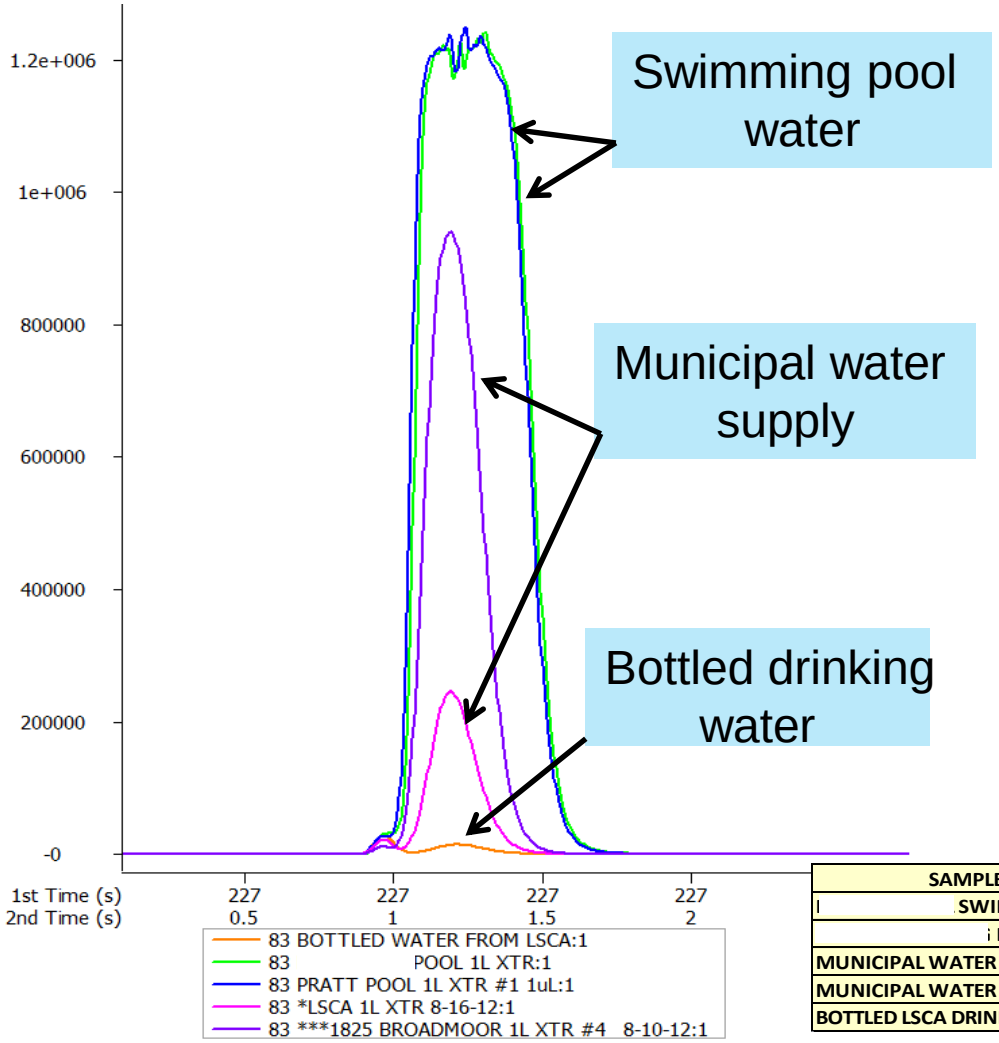
VOCs and Miscellaneous DBPs

Benzene, 1,4-dichloro-

CHLOROFORM CALIBRATION



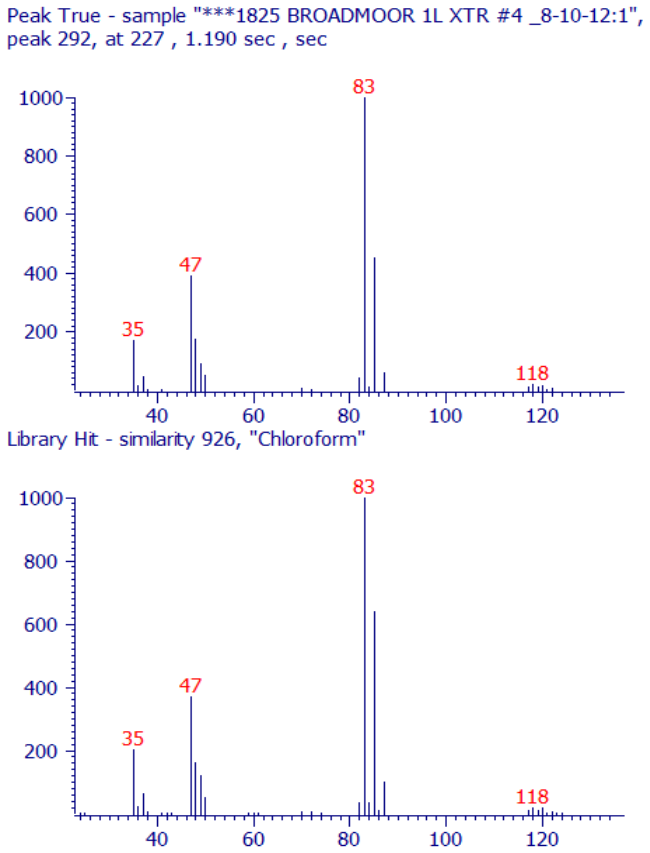
PEAK AREA COMPARISON of CHLOROFORM: MUNICIPAL WATER SUPPLY vs. POOL WATER vs. BOTTLED DRINKING WATER



Swimming pool water

Municipal water supply

Bottled drinking water



SAMPLES	Name ID	Similarity	R.T. (s)	Area	Height	Library
SWIMMING POOL	Chloroform	941	227 , 1.230	140687824	2631096	Wiley9
POOL	Chloroform	888	227 , 1.185	77166100	1781443	Wiley9
MUNICIPAL WATER 1825 BROAD	Chloroform	926	227 , 1.190	40700362	940928	Wiley9
MUNICIPAL WATER LSCA	Chloroform	868	227 , 1.190	10550115	284422	Wiley9
BOTTLED LSCA DRINKING WATER	Chloroform	890	227 , 1.215	646089	18007	replib

Conclusions from the Study

- These SPE-GCxGC-TOFMS analyses identified 37 DBPs by comparison to EPA lists including Method 551.1 and the EPA 2005 DBP National Occurrence Study. Further data review compared to the TEDX list of EDCs found ten chemicals which are known endocrine-disrupting compounds.

- This work emphasizes the need for instrumentation that will detect and identify sources of long-term environmental exposure to DBPs and EDCs that can lead to ecological destruction and serious health effects. The application of GCxGC-TOFMS for this work presents a sensitive and robust instrumental option for the detection of DBPs and EDCs, as well as other untargeted contaminants in treated water.

Conclusions from both studies

- A robust method using SPE-GCxGC-TOFMS for the analysis of EDCs and DBP in water was demonstrated
- 102 EDCs or miscellaneous pollutants were found in this study of the watershed
- Over 30 DBPs were identified in potable and swimming pool water
- Part per trillion (ppt) detectability of EDCs was accomplished for selected analytes using GCXGC-TOFMS

For More Information

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