SIMULTANEOUS ANALYSIS FOR DIOXINS, PCBS AND PBDES WITH A FULLY AUTOMATED SAMPLE PREPARATION SYSTEM (II: VALIDATION)

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If you want to see the system, go to the Booth 1 & 2.

INTRODUCTION

Poster code 2,1016

The unique automated sample preparation system, GO-HT series, have been on the European market since 2015. The system is authorized as a validated method by some European accreditation laboratories. In the same way, the EU commission has adopted legislation to reduce or halt the sale and use of certain brominated flame retardants (BFRs) in order to protect human health and the environment. The EFSA recommended the monitoring of BFRs in foodstuffs based on commission recommendation 2014/118. Therefore, the demand for viable technologies for the simultaneous analysis of PCDDs/PCDFs, PCBs, and PBDEs in foodstuffs has increased in European laboratories. We previously reported a procedure for the simultaneous analysis for PCDDs/PCDFs, PCBs, and PBDEs using the system. The method is much more effective than manual purification steps, which involve complicated operations. In this presentation, we report the simultaneous analysis of PCDDs / PCDFs / PCBs / PBDEs in feed and food samples as an application of the developed system. Based on the results of the quality control samples, real samples (unknowns), procedure blanks, and standard reference materials (SRMs), the fractionations of PCDDs / PCDFs / PCBs / PBDEs, limit of quantification (LOQ), accuracy, and recoveries are discussed.

CONCLUSIONS

<u>The ways of efficient elution of PBDEs from Si-column: following two methods,</u>

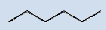
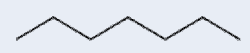
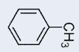
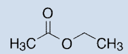
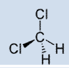
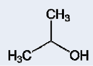
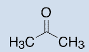
1. Adding small amount of Ethyl acetate to the sample solution, However, it depends on the sample species and sample volume / weight. So, this way is helpful for the laboratories that usually implement the inspection of the common foodstuff products.
2. Increasing elution volume, 140 ~ 160 mL of n-hexane at 60 dC. It is not affected by the sample species, etc. So, this method is effective for the laboratories that usually treat the many sample species.

<u>The background level of column and system of 209-decaBDE is extremely near zero.</u>

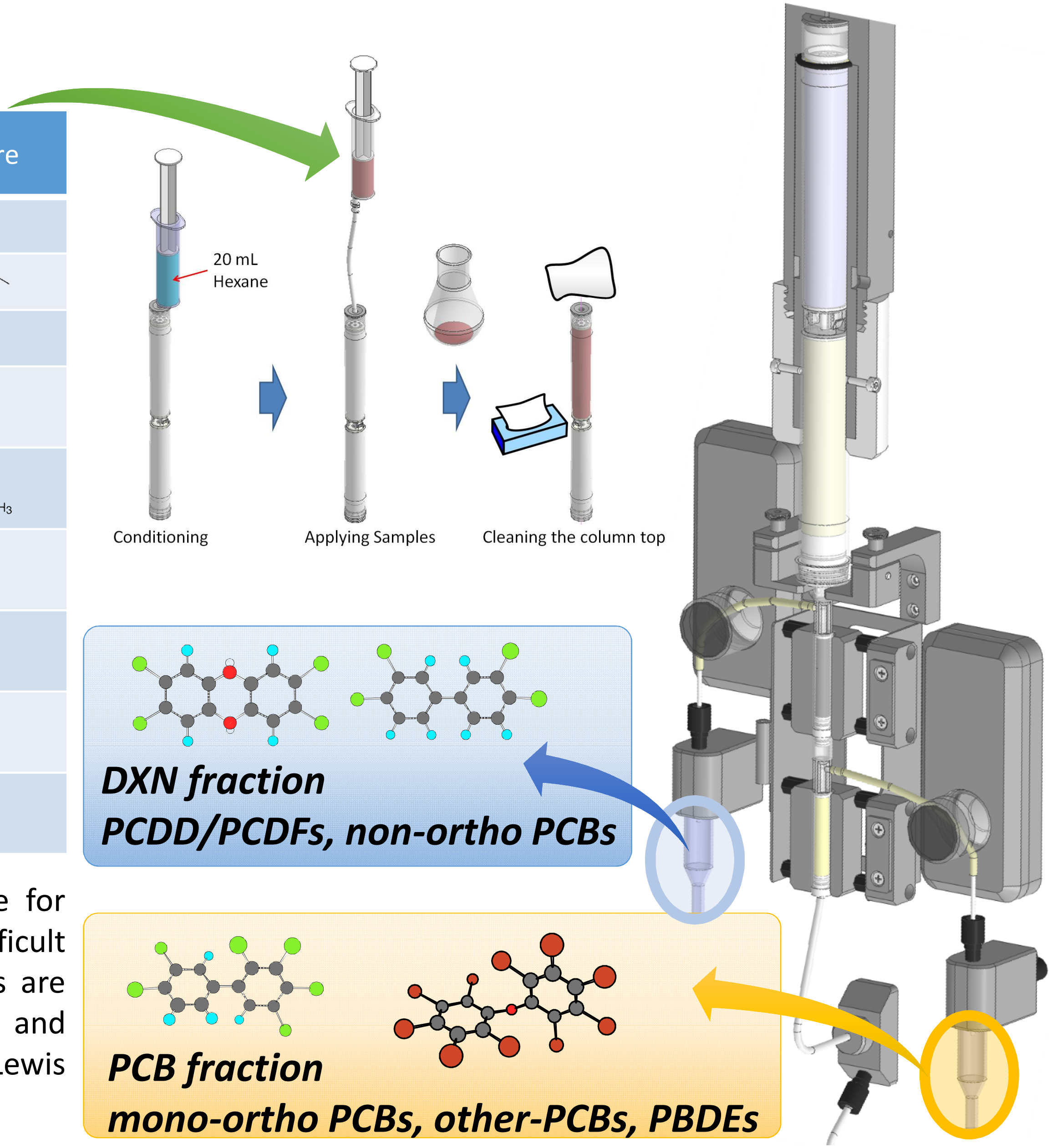
<u>The recoveries and trueness of PCDDs/PCDFs and PCBs were obtained good results to all the samples.</u>

<u>Trueness of PBDEs obtained by use of CRM is within ±20%.</u>

SAMPLE PREPARATION

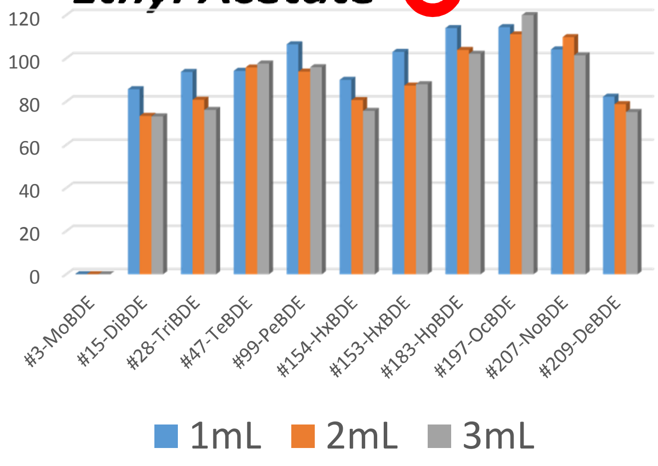
Mobile Phase	Additive	Polarity	Boiling point dC	Structure
n-Hexane	---	1.88	69	
n-Heptane	---	1.92	98	
Cyclohexane	---	2.02	80	
n-Hexane	Toluene	2.38	110	
	Ethyl acetate	6.02	76	
	THF	7.58	65	
	DCM	8.93	40	
	2-propanol	20.33	98	
	Acetone	20.70	56	

The normal method requires 90 mL of n-Hexane as mobile phase for elution of PCDDs/PCDFs and PCBs from Si-column. However, It is difficult to elute PBDEs from Si-column with only n-hexane, because PBDEs are strongly adsorbed on the silica gel because of higher polarities and stronger interaction that occurs between a halogen (bromine) atom (Lewis acid) and a Lewis base (silica gel). (Halogen bond: F < Cl < Br < I)

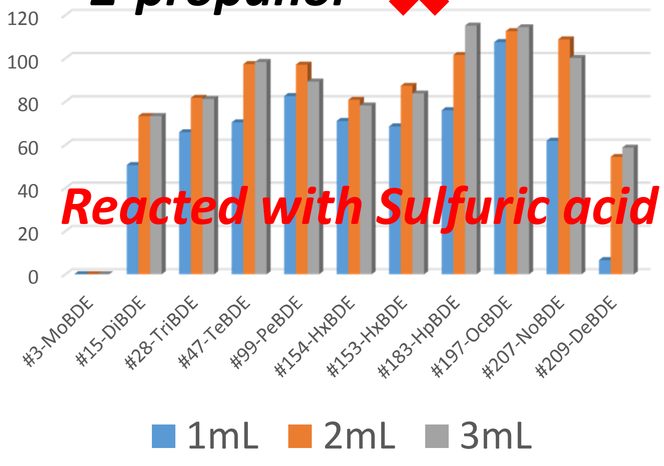


Recoveries of PBDEs in the PCB fraction (When using Additive eluents)

Ethyl Acetate ○

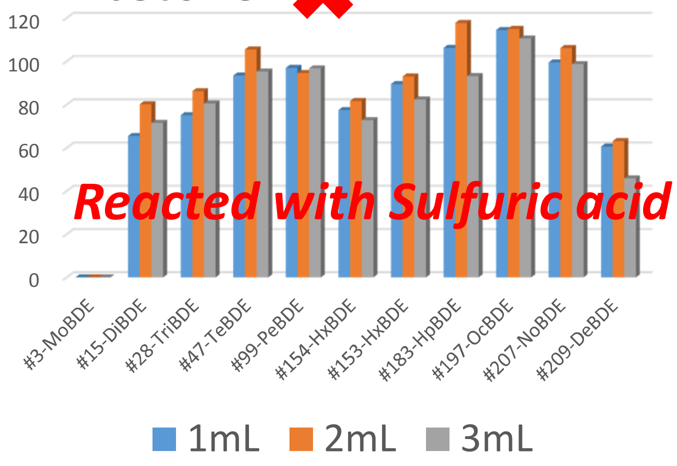


2-propanol ✖

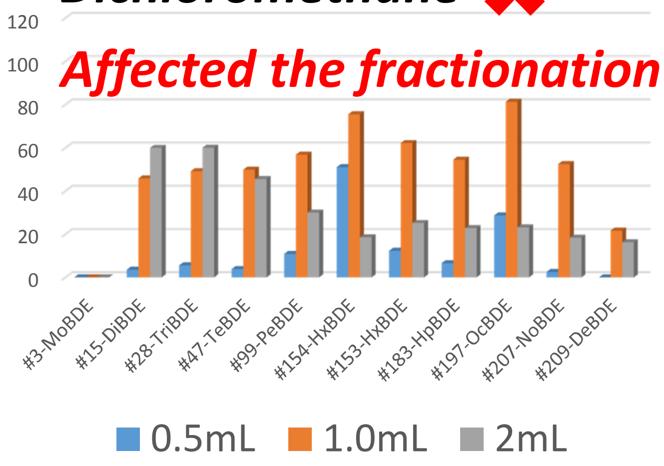


THF ✖

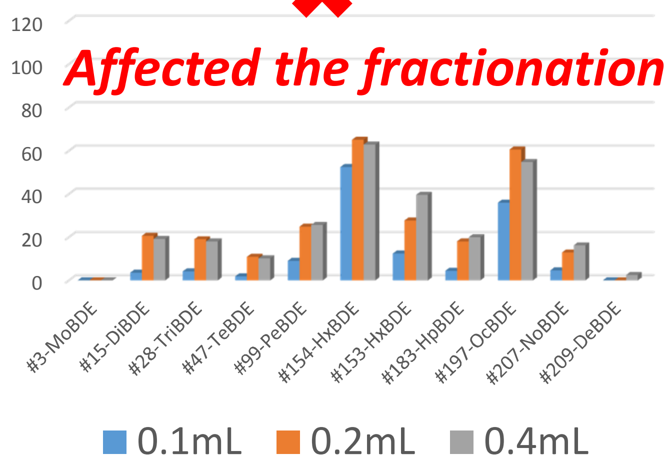
Acetone ✖



Dichloromethane ✖



Toluene ✖



Recoveries of PBDEs in the n-hexane eluted from only Multi-layer silica gel column

	60dC Hex +1 mL EtAc			60dC Hex				Room Temp Hex + 1 mL EtAc				Room Temp Hex					
	70mL	80mL	90mL	90mL	120mL	140mL	160mL	90mL	120mL	140mL	160mL	90mL	120mL	140mL	160mL	180mL	200mL
#3-MoBDE	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%	0%
#7-DiBDE	0%	0%	0%	0%	0%	0%	0%	0%	6%	6%	5%	0%	0%	0%	0%	0%	0%
#15-DiBDE	102%	105%	107%	11%	77%	104%	104%	48%	96%	97%	105%	0%	0%	0%	0%	10%	91%
#17-TriBDE	53%	51%	46%	0%	0%	25%	39%	14%	75%	86%	86%	0%	0%	0%	0%	0%	0%
#28-TriBDE	103%	99%	101%	10%	70%	100%	102%	57%	92%	99%	99%	0%	0%	0%	0%	4%	59%
#49-TeBDE	103%	107%	100%	1%	62%	100%	100%	49%	90%	100%	98%	0%	0%	0%	0%	1%	18%
#71-TeBDE	113%	106%	101%	0%	44%	97%	110%	43%	91%	102%	105%	0%	0%	0%	0%	0%	5%
#47-TeBDE	103%	109%	102%	3%	70%	107%	110%	58%	95%	104%	108%	0%	0%	0%	0%	1%	25%
#66-TeBDE	100%	106%	99%	6%	66%	99%	108%	59%	97%	108%	105%	0%	0%	0%	0%	2%	33%
#77-TeBDE	97%	107%	103%	2%	68%	105%	110%	61%	101%	107%	113%	0%	0%	0%	0%	1%	26%
#100-PeBDE	102%	107%	105%	43%	83%	97%	107%	84%	92%	98%	98%	0%	0%	1%	4%	34%	91%
#119-PeBDE	101%	110%	109%	50%	91%	95%	107%	88%	99%	101%	98%	0%	0%	3%	12%	68%	93%
#99-PeBDE	104%	109%	110%	8%	67%	102%	103%	66%	95%	102%	102%	0%	0%	0%	0%	2%	30%
#85-PeBDE	96%	102%	99%	0%	0%	60%	88%	34%	86%	97%	97%	0%	0%	0%	0%	0%	0%
#126-PeBDE	101%	110%	103%	8%	70%	102%	112%	70%	103%	104%	110%	0%	0%	0%	0%	2%	34%
#154-HxBDE	108%	109%	107%	47%	94%	108%	109%	88%	98%	107%	104%	0%	1%	2%	6%	46%	96%
#153-HxBDE	113%	108%	107%	8%	75%	106%	111%	69%	96%	107%	105%	0%	0%	0%	0%	1%	29%
#138-HxBDE	101%	102%	103%	0%	0%	63%	96%	39%	86%	95%	104%	0%	0%	0%	0%	0%	0%
#156-HxBDE	104%	110%	108%	0%	33%	88%	110%	51%	99%	110%	123%	0%	0%	0%	0%	0%	1%
#184-HpBDE	118%	91%	117%	70%	104%	101%	117%	102%	109%	114%	118%	0%	0%	6%	23%	83%	90%
#183-HpBDE	106%	97%	100%	2%	61%	86%	95%	60%	89%	107%	100%	0%	0%	0%	0%	1%	17%
#191-HpBDE	110%	106%	82%	5%	67%	91%	98%	74%	108%	122%	113%	0%	0%	0%	0%	2%	28%
#197-OcBDE	109%	113%	99%	8%	69%	98%	105%	75%	93%	98%	107%	0%	0%	0%	0%	1%	22%
#196-OcBDE	117%	119%	108%	0%	17%	77%	102%	48%	92%	96%	101%	0%	0%	0%	0%	0%	1%
#207-NoBDE	109%	104%	117%	0%	50%	106%	112%	65%	95%	112%	108%	0%	0%	0%	0%	0%	3%
#206-NoBDE	110%	113%	116%	0%	0%	53%	89%	47%	93%	112%	112%	0%	0%	0%	0%	0%	0%
#209-DeBDE	100%	123%	106%	0%	0%	71%	95%	59%	77%	91%	99%	0%	0%	0%	0%	0%	0%
Required Vol: 70 mL				***140~160mL***				***140~160mL***				***>>200mL***					

Recoveries of PBDEs in the samples

13C-labeled PBDEs (no adding ethyl acetate)

	Pig fat	RapeSeed	Rice	Olive	Sesame	Soybean	Palm	Sunflower		Mineral		Raw milk fat			Butter		
	3g	3g	3g	3g	3g	3g	3g	3g	2g	0.5g	1.0g	2.5g	2.0g	1.5g	2.5g	2.0g	1.5g
#3-MoBDE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
#15-DiBDE	85.7	92.5	68.3	90.6	75.4	71.0	82.7	84.3	86.8	0.0	0.0	114.7	97.2	70.1	85.2	80.9	21.8
#28-TriBDE	80.7	96.1	61.7	83.3	75.4	69.8	85.6	83.2	87.0	0.0	0.0	107.8	94.6	77.0	90.7	86.5	21.6
#47-TeBDE	94.3	85.0	78.0	77.3	80.3	77.3	86.1	78.4	86.1	0.0	0.0	92.4	79.0	75.0	81.0	62.0	4.6
#99-PeBDE	82.4	96.7	78.2	66.6	74.8	67.0	102.2	85.1	84.7	0.0	0.0	84.2	78.2	61.5	81.9	68.1	11.0
#154-HxBDE	95.7	86.7	69.6	75.8	98.2	79.0	81.3	78.0	86.5	0.0	0.0	57.6	88.7	69.8	93.9	93.6	75.6
#153-HxBDE	87.5	77.9	74.8	78.9	92.0	67.2	88.8	77.9	82.1	0.0	0.0	62.8	66.7	58.4	67.3	61.5	7.8
#183-HpBDE	98.1	78.0	77.8	81.7	89.0	76.6	86.9	73.4	82.7	0.0	0.0	83.5	81.9	72.1	85.0	53.8	3.7
#197-OcBDE	128.7	117.8	103.5	122.8	111.5	89.8	120.8	91.7	109.5	0.0	0.0	125.1	126.6	99.6	109.9	99.0	5.2
#207-NoBDE	84.8	80.7	76.6	70.8	76.2	70.6	104.0	76.2	90.3	0.0	0.0	107.3	61.7	72.7	76.8	6.6	0.0
#209-DeBDE	28.4	34.9	50.4	25.7	42.4	34.4	34.1	44.2	56.4	0.0	0.0	32.2	0.0	0.0	0.0	0.0	0.0

	1 Compound Feed for fish			2 Compound Feed for fish			3 Compound Feed for fish			Fish liver oil			Fish Oil		
	3g	2g	1g	3g	2g	1g	3g	2g	1g	3g	2.5g	2g	2g	1.5g	1g
#3-MoBDE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
#15-DiBDE	82.4	49.9	0.0	54.4	48.5	0.0	93.6	86.0	1.0	53.9	68.8	54.3	78.2	43.1	0.0
#28-TriBDE	90.0	64.8	0.0	59.3	51.7	0.0	97.8	91.4	0.0	73.1	81.7	70.6	91.5	44.4	0.0
#47-TeBDE	86.0	50.0	0.0	63.3	47.2	0.0	97.7	69.8	0.0	92.8	89.4	78.6	86.1	9.0	0.0
#99-PeBDE	77.2	72.0	0.0	83.1	54.8	0.0	88.6	76.9	0.0	127.6	73.0	83.2	78.0	22.0	0.0
#154-HxBDE	88.8	59.4	0.0	63.2	57.8	0.0	80.5	78.6	13.4	60.9	74.3	67.8	79.2	67.3	3.7
#153-HxBDE	66.5	50.7	0.0	60.9	42.5	0.0	72.0	60.1	0.0	68.6	69.5	52.1	62.1	16.3	0.0
#183-HpBDE	84.6	59.1	0.0	72.4	56.3	0.0	102.6	51.6	0.0	89.6	68.4	46.0	57.6	0.0	0.0
#197-OcBDE	110.4	125.8	0.0	89.9	74.0	0.0	126.6	103.5	0.0	126.1	120.5	125.4	103.6	0.0	0.0
#207-NoBDE	92.7	23.9	0.0	64.4	36.9	0.0	109.5	6.7	0.0	95.6	69.8	0.0	20.9	0.0	0.0
#209-DeBDE	46.3	0.0	0.0	20.1	0.0	0.0	59.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

	Salmon			Egg fat				Poultry			Poultry liver fat			Bovine		
	3g	2.5g	2g	3g	2.5g	2.0g	1.5g	3g	2.5g	2.0g	1.5g	1g	0.5g	2.5g	2.0g	1.5g
#3-MoBDE	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
#15-DiBDE	109.4	75.9	72.2	84.6	54.0	47.0	60.3	69.9	118.3	71.7	10.2	0.0	0.0	77.8	80.9	67.6
#28-TriBDE	99.6	80.4	79.4	87.5	63.3	56.8	69.0	72.8	107.5	80.1	11.2	0.0	0.0	80.3	81.3	75.4
#47-TeBDE	90.7	95.8	84.7	95.4	74.8	62.6	79.8	88.1	96.6	91.7	3.6	0.0	0.0	89.6	61.4	73.2
#99-PeBDE	86.9	80.2	70.1	87.6	65.8	55.3	69.3	82.2	70.6	74.8	7.1	0.0	0.0	77.1	80.4	68.5
#154-HxBDE	107.8	105.5	110.7	100.7	103.9	108.7	103.4	94.3	98.1	101.8	54.2	0.0	0.0	98.1	86.8	96.4
#153-HxBDE	90.2	96.0	96.8	95.2	78.0	66.6	73.8	89.2	82.6	86.2	9.0	0.0	0.0	80.5	68.3	78.5
#183-HpBDE	78.4	94.4	83.2	91.6	82.9	70.3	77.7	97.2	74.3	83.2	5.6	0.0	0.0	88.2	60.9	69.9
#197-OcBDE	103.8	132.2	120.2	119.7	94.5	113.9	114.6	104.5	115.2	104.6	14.6	0.0	0.0	120.0	119.2	98.6
#207-NoBDE	90.0	94.7	85.9	84.7	66.2	72.7	82.0	77.7	96.6	79.5	0.0	0.0	0.0	89.8	53.2	36.4
#209-DeBDE	31.6	43.5	18.5	45.2	34.9	29.1	40.0	35.4	28.4	24.3	0.0	0.0	0.0	33.0	0.0	0.0

The recoveries of #209-BDE of all the samples are lower. This indicates that it is difficult to elute #209-BDE from Si-column with only 90 mL of n-hexane. Notably, the recoveries of the following samples are not very good, less than 1 to 2 g of Mineral oil, raw milk fat, butter, some feeds, fish liver oil, fish oil, poultry liver fat, and bovine (beef meat) fat.

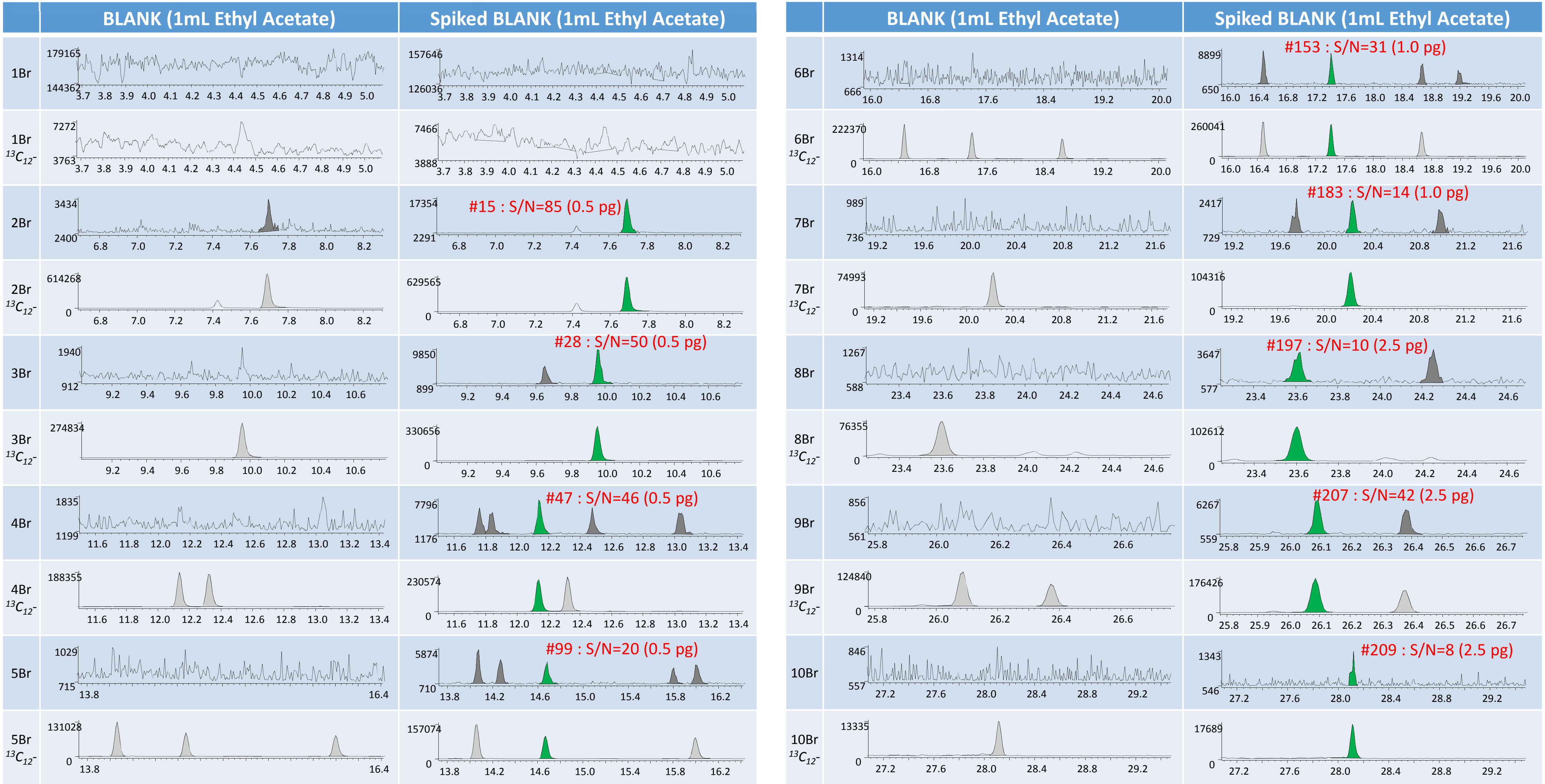
Native PBDEs

	Milk fat 2.5g			Butter 2.5g		
Hexane	140mL	90mL	90mL	140mL	90mL	90mL
Ethyl Acetate	---	0.2mL	0.4mL	---	0.2mL	0.4mL
#3-MoBDE	0%	0%	0%	0%	0%	0%
#7-DiBDE	0%	0%	0%	0%	0%	0%
#15-DiBDE	116%	101%	79%	102%	100%	107%
#17-TriBDE	66%	68%	7%	54%	52%	68%
#28-TriBDE	118%	112%	75%	107%	106%	111%
#49-TeBDE	106%	109%	59%	97%	97%	103%
#71-TeBDE	106%	108%	67%	97%	94%	109%
#47-TeBDE	119%	121%	86%	101%	97%	104%
#66-TeBDE	112%	106%	67%	103%	88%	97%
#77-TeBDE	108%	88%	64%	94%	64%	73%
#100-PeBDE	102%	115%	84%	105%	100%	99%
#119-PeBDE	102%	116%	80%	108%	104%	105%
#99-PeBDE	111%	113%	85%	104%	98%	100%
#85-PeBDE	105%	119%	81%	110%	86%	92%
#126-PeBDE	101%	94%	68%	105%	72%	65%
#154-HxBDE	117%	105%	81%	104%	97%	107%
#153-HxBDE	118%	105%	83%	109%	100%	98%
#138-HxBDE	115%	99%	86%	118%	88%	101%
#156-HxBDE	124%	82%	69%	100%	69%	79%
#184-HpBDE	90%	114%	72%	83%	73%	79%
#183-HpBDE	106%	120%	79%	88%	104%	115%
#191-HpBDE	94%	119%	118%	120%	118%	116%
#197-OcBDE	107%	95%	74%	91%	83%	91%
#196-OcBDE	103%	85%	72%	77%	82%	94%
#207-NoBDE	108%	106%	72%	99%	82%	98%
#206-NoBDE	115%	119%	84%	102%	75%	116%
#209-DeBDE	107%	118%	61%	90%	82%	85%

The recoveries of #209-BDE obtained by improved two methods seem to be better than normal method. However, The way of addition of ethyl acetate should be valided one step further to each sample.

CHROMATOGRAMS OF PROCEDURE BLANK

(BD-5HT, 15m, 1 injection measurement)



VALIDATION: RESULTS OF SRM

Cod liver oil 1 g (CIL, EDF-5463)

	Reference Value		2015's Validation (n=4)				2016's Validation (n=4)					
Compounds	Assigned	SD x2	Mean	SD	RSD_R	Deviation	Mean	SD	RSD_R	Deviation	LOD SDx3	LOQ SDx10
Congeners	pg/g fat		pg/g fat		%	%	pg/g fat		%	%	pg/g	
#15-DiBDE	-		(2.1)	0.21	10		ND				1.6	5.4
#17-TriBDE	8.83	± 5.76	(6.6)	0.94	14	-25	(8.4)	0.46	5	-5	2.7	9.0
#28-TriBDE	40.1	± 13.9	37.9	1.54	4	-5	38.3	1.36	4	-5	1.1	3.8
#49-TeBDE	-		278.8	8.84	3		260.3	7.09	3		2.1	6.9
#47-TeBDE	1480	± 480	1499.2	51.84	3	1	1497.4	23.07	2	1	2.2	7.2
#66-TeBDE	48.4	± 29.4	50.1	6.70	13	3	49.5	1.66	3	2	2.0	6.8
#77-TeBDE	-		ND				(5.9)	0.43	7		3.4	11.4
#100-PeBDE	357	± 50.6	350.7	18.62	5	-2	372.3	19.22	5	4	2.0	6.6
#119-PeBDE	-		26.8	4.42	16		33.1	2.18	7		5.1	16.9
#99-PeBDE	193	± 70.4	160.6	4.93	3	-17	165.4	4.47	3	-14	3.1	10.3
#154-HxBDE	229	± 88.8	207.2	7.41	4	-10	233.4	9.81	4	2	5.4	18.0
#153-HxBDE	33.9	± 6.64	32.1	2.13	7	-5	35.0	3.16	9	3	2.6	8.7
#183-HpBDE	-		(11.4)	0.84	7		ND				5.9	19.7
#207-NoBDE	-		(24.0)	2.37	10		(17.9)	2.48	14		8.8	29.4
#206-NoBDE	-		(30.6)	3.53	12		(16.7)	2.87	17		12.9	43.0
#209-DeBDE	-		131.3	21.65	16		134.1	13.54	10		35.5	118.3
Labeled standard Recovery												
#15-DiBDE			79.8	1.9	2.4		81.3	3.4	4.2			
#28-TriBDE			84.1	6.8	8.1		91.7	2.4	2.7			
#47-TeBDE			78.1	25.0	32.0		91.4	2.2	2.4			
#99-PeBDE			92.2	24.6	26.6		90.5	1.4	1.5			
#154-HxBDE			81.8	17.4	21.2		77.7	3.7	4.8			
#153-HxBDE			88.5	19.1	21.6		73.7	1.8	2.4			
#183-HpBDE			95.8	10.4	10.8		70.3	3.6	5.1			
#197-OcBDE			87.0	22.8	26.3		105.5	6.3	5.9			
#207-NoBDE			79.5	13.4	16.9		79.9	1.9	2.4			
#209-DeBDE			68.2	16.6	24.4		52.8	1.3	2.4			