Advances in distinguishing groundwater influenced by Oil Sands Process-affected Water (OSPW) from natural bitumen-influenced groundwaters.

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Supporting Information

Photos of Field Work

Figure S1 – Photos of shallow groundwater sampling at the edge of the Athabasca River, adjacent to Tar Island Dyke (Pond 1), using the mini-profiler system.

Figure S2 – Photos of shallow groundwater sampling at the edge of the Athabasca River or one of its tributaries away from any tailings pond (i.e., background samples) using the mini-profiler system.

Figure S3 – Photo showing small globules of bitumen extracted with shallow groundwater using the mini-profiler, collected in a plastic volumetric flask, which occurred at several of the background sites.

Table S1. Details on sample collection; those with similar collection details are grouped.

Collected Sample	**Container**	**Preservation**
Major anions (chloride, nitrate, nitrite, sulphate, phosphate); Artificial Sweeteners	30mL HDPE	Filtered (0.45 µm)
Major cations; metals (+ metalloids)	250 mL HDPE	filtered; pH <2, with 10% nitric acid
Ammonium	30mL HDPE	filtered; pH 5-6, with 10% hydrochloric acid

PFAS Analysis

Briefly, 300 ml to 500 ml samples of groundwater were spiked with 30 µL of 6 to 15 ng ml^{-1} methanolic mixture of isotopically labeled surrogates (to track extraction efficiency) and adjusted to pH 3 using formic acid. After conditioning 150 mg SPE (OASIS WAX, Waters) using methanol and SPE-polished water, samples were loaded at a rate of 1 ml min^{-1}. PFAS were eluted using 5 ml of 1% ammonia in methanol and concentrated to just dryness using a gentle stream of nitrogen gas. Residue was reconstituted in 0.5 ml of 1:1 water/methanol and spiked with 30 µL of a separate isotopically labeled standard cocktail (6 to 15 ng ml^{-1}) to monitor for matrix effects. PFAS analysis was by ultrahigh performance liquid chromatography tandem mass spectrometry (UHPLC-MS/MS, Waters XEVO TQS system) operated in negative electrospray ionization mode using previously reported instrumental parameters [50] with further details provided (Tables S2-4). OSPW samples were processed using analogous methods but using 50 ml sample extractions due to the high concentrations. Each analyte was quantified using relative response to its corresponding isotopically labeled standard via a 15-level calibration curve ranging from 0.003-0.002 ng ml^{-1} to 25 -30 ng ml^{-1}. Method blanks were processed alongside samples to ascertain background contamination and spike and recovery experiments were conducted to determine extraction efficiency and matrix effects. All PFAS were below detection limits (<LOD) in method blanks with the exception of perfluorooctanoate which ranged from 0.011 to 0.020 ng ml^{-1} in the final 0.5 ml extract.

Extraction efficiency (EE) was calculated using:

$$\boldsymbol{EE} = \frac{\boldsymbol{A_{pre-extraction\ surrogate}}}{\boldsymbol{A_{post-extraction\ surrogate}}} \times \mathbf{100}\%$$

Where $A_{pre\text{-}extraction}$ is the peak area of the surrogate that was spiked into the sample before extraction (for example $^{13}C_{1,2,3,4}$-PFOS) and $A_{post\text{-}extraction}$ is the peak area of the surrogate that was spiked into the sample after extraction (i.e. $^{13}C_{1,2,3,4,5,6,7,8}$-PFOS) just before instrumental analysis. The measure of extraction efficiency provides an indication of the extraction recovery without influence of the matrix enhancement or suppression since both numerator and denominator contain the same matrix. An ideal extraction method produces EE close to 100%.

Matrix Recovery (MR) was calculated using:

$$\boldsymbol{MR} = \frac{\boldsymbol{A_{post-extraction\ surrogate}}}{\boldsymbol{A_{calibration\ standard}}} \times \mathbf{100}\%$$

Where $A_{post\text{-}extraction}$ is the peak area of the surrogate that was spiked into the sample after extraction (i.e. $^{13}C_{1,2,3,4,5,6,7,8}$-PFOS) just before instrumental analysis and $A_{calibration\ standard}$ is the peak area of the same surrogate at equivalent concentration in solvent only (no matrix). The measure of matrix surrogate recovery provides an indication of the matrix effect without influence of analyte recovery because surrogate in the numerator was spiked into the matrix after extraction. When MR <100%, matrix suppression is occurring and >100% suggests matrix enhancement of the analyte signal. An ideal extraction method produces MR close to 100%.

Table S2. Average ± standard error recovery (%) of isotopically labeled standards in samples using peak area. Extraction efficiency is calculated using the surrogate spiked prior to extraction and compared to the surrogate spiked post-extraction. Matrix effects were evaluated by comparing the recovery of the surrogate added to the extract to a solvent standard.

Extraction efficiency (%), based on surrogate spiked to sample before extraction		**Recovery of Matrix spike (post-extraction) (%)**	
$^{13}C_{1,2,3,4}$-PFOS	$^{18}O_{1,2}$-PFHxS	$^{13}C_{1,2,3,4,5,6,7,8}$-PFOS	$^{13}C_{1,2,3}$-PFHxS
86 ±5.0	87 ±4.3	99 ±1.2	98 ±2.6

Table S3. Limit of quantitation (LOQ) for PFAS in OSPW and groundwater. Instrument detection limits are based on concentration corresponding to signal to noise of 5 using solvent standard. Matrix-specific limits of quantitation are in units of ng/L corresponding to sample extraction volume (500 ml groundwater and 0.5 ml extract; 50 ml OSPW and 0.5 ml extract). *LOQ for PFOA in OSPW and groundwater is based on average PFOA concentration in blanks (0.0155 ng/ml).

Analyte	**Instrument detection limits (ng/ml)**	**LOQ in OSPW (ng/L)**	**LOQ in groundwater (ng/L)**
PFHxA	0.002	0.02	0.002
PFHpA	0.003	0.03	0.003
PFOA	0.004	0.15*	0.016*
PFNA	0.004	0.04	0.004
PFDA	0.004	0.04	0.004
PFUnDA	0.004	0.04	0.004
PFDoA	0.004	0.04	0.004
PFBS	0.003	0.03	0.003
PFHxS	0.002	0.02	0.002
PFOS	0.002	0.02	0.002

Table S4. Instrument parameters for PFAS analysis

Liquid Chromatograph				
Instrument name	Acquity UPLC I class (Waters Corporation)			
Injection volume	2 µL			
Column	BEH C-18, 2.1 x 100 mm, 1.7 µm			
Column temperature	40 °C			
Mobile Phase	A: 0.1 mM ammonium acetate (>98%, Sigma Aldrich) in SPE-cleaned water (Optima Grade, Fisher Scientific) B: methanol (Optima Grade, Fisher Scientific)			
Gradient elution	**Time (min)**	**Flow Rate, ml min^{-1}**	**%A**	**%B**
	0	0.4	75	25
	0.5	0.4	75	25
	5.0	0.4	15	85
	5.1	0.4	0	100
	5.6	0.4	0	100
	7.0	0.55	0	100
	9.0	0.4	75	25
	13.0	0.4	75	25

Mass spectrometer	
Instrument name	XEVO TQ-S (Waters Corporation)
Ionization mode	Electrospray negative ionization
Detection mode	Multiple reaction monitoring (MRM)
Source temperature	150 °C
Capillary voltage	0.6 kV
Desolvation gas temperature	450 °C
Collision gas flow rate	800 L hr^{-1}

MRM precursor to product ion transitions:

Analyte	MRM (m/z)	Cone (V)	Collision Energy (V)
PFHxA	313→269, 119	16	10, 17
PFHpA	363→319, 169	16	10, 18
PFOA	413→369, 169	16	11, 18
PFNA	463→419, 219	10	10, 17
PFDA	513→469, 219	10	10, 17
PFUnDA	563→519, 269	10	10, 17
PFDoA	613→569, 169	8	12, 28
PFBS	299→ 99, 80	6	30, 30
PFHxS	399→99, 80	6	32, 32
PFOS	499→ 99, 80	2	36, 40

Table S5. Tests for equality between Background GW and OSPW groups.
Median tests done on untransformed values; t-tests on transformed values [$\log(x+x_{min}) - \log(x_{min})$]

	Median Test		T-test	
Variable	**X^2**	**P**	**T**	**P**
∑ Family A	5.00	0.025	-15.35	<0.001
∑ Family B	5.00	0.025	-8.29	<0.001
Family A1-6	5.00	0.025	-14.76	<0.001
A-1	7.50	0.006	-28.30	<0.001
A-2	5.00	0.025	-14.78	<0.001
A-3	11.67	0.001	-30.79	<0.001
A-4	7.50	0.006	-20.85	<0.001
A-5	11.67	0.001	-37.27	<0.001
A-6	5.00	0.025	-14.33	<0.001
A-7	5.00	0.025	-15.53	<0.001
A-8	5.00	0.025	-14.27	<0.001
B-1	5.00	0.025	-9.36	<0.001
B-2	5.00	0.025	-8.20	<0.001
O_2:O_4	3.96	0.047	-2.96	0.016

Table S6. Family A and B acids, as methyl esters, in OSPW and Background groundwaters sampled in this study. <DL denotes concentrations below detection limit, 0.2 µg/L.

	Family A/B Isomer Concentration (ug/L)									
OSPW	**A-1**	**A-2**	**A-3**	**A-4**	**A-5**	**A-6**	**A-7**	**A-8**	**B-1**	**B-2**
MLSB	16.7	25.0	20.2	12.4	10.3	34.6	6.5	5.2	32.9	24.5
NVS_2_MLSB_1	19.6	53.7	24.5	16.7	10.0	75.1	8.9	8.8	108.3	105.9
NVS_2_MLSB_2	19.0	57.1	28.2	17.6	11.8	78.1	8.7	6.7	107.9	94.1
NVS_2_MLSB_3	19.6	54.7	25.3	14.6	9.4	70.4	8.3	8.9	101.1	112.6
NVS_2_MSB_4	18.7	52.0	23.2	14.1	14.1	66.6	7.9	8.4	98.5	101.9
NVS_2_MLSB_4.1	18.5	46.9	22.3	13.3	8.3	62.4	6.3	4.6	87.3	79.4
NVS_2_MLSB_4.2	16.0	41.2	22.6	13.6	8.9	57.4	7.9	8.0	26.1	79.3
NVS_2_MLSB_5	15.2	43.2	20.7	12.6	9.1	56.7	7.5	7.8	74.8	94.7
NVS_2_MLSB_5.1	16.3	44.1	22.4	5.2	9.1	57.2	7.0	8.0	87.8	72.7
NVS_MLSB_5.2	9.8	18.7	8.5	5.2	4.0	16.9	3.6	0.0	43.6	35.2
NVS_1_SWIP_1	23.9	76.3	33.8	24.5	11.8	108.3	17.2	18.9	129.0	105.8
NVS_1_SWIP_2	26.4	74.9	33.5	21.3	14.8	99.7	15.8	16.5	95.1	159.2
NVS_1_SWIP_3	23.2	80.1	39.2	26.2	17.2	114.5	17.9	19.1	117.8	132.6
NVS_1_SWIP_4	26.4	79.5	36.6	24.8	15.7	111.4	18.9	18.9	120.7	109.0
NVS_1_SWIP_4.1	26.1	79.9	37.0	24.9	15.3	109.3	18.1	18.8	129.2	146.7
NVS_1_SWIP_4.2	21.9	63.9	32.6	20.9	12.7	87.3	11.9	12.5	114.0	93.3
NVS_1_SWIP_5	26.5	89.1	45.3	29.8	21.0	128.7	21.4	22.0	37.9	125.4
NVS_1_SWIP_5.1	23.4	79.0	37.1	24.6	18.7	117.9	19.8	20.4	126.3	116.4
NVS_1_SWIP_5.2	18.0	38.2	17.8	11.1	5.1	40.9	4.6	0.0	84.8	66.3
NVS_3_SWSS_1	24.2	56.0	31.3	17.8	8.9	75.7	11.1	10.5	105.1	99.9
NVS_3_SWSS_2	22.9	57.2	31.6	17.7	10.8	74.8	10.5	11.2	108.7	86.3
NVS_3_SWSS_3	21.7	51.0	29.0	16.8	9.3	70.1	9.7	10.2	106.7	80.5
Background Groundwater	**A-1**	**A-2**	**A-3**	**A-4**	**A-5**	**A-6**	**A-7**	**A-8**	**B-1**	**B-2**
DP 1	<DL	2.2	<DL	0.4	<DL	2.0	0.4	0.4	4.9	4.9
DP 2	<DL	<DL	<DL	<DL	<DL	<DL	0.6	<DL	10.1	12.8
DP 3	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL
BG-1	<DL	<DL	<DL	<DL	<DL	<DL	<DL	0.1	<DL	<DL
BG-2	<DL	0.9	<DL	0.3	<DL	0.5	<DL	<DL	1.4	2.9
BG-3	0.4	1.3	0.5	0.4	0.1	0.7	0.1	0.1	3.4	3.4

BG-4	<DL	0.2	<DL	<DL	<DL	0.1	0.1	<DL	0.4	0.5
BG-5	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL
BG-6	<DL	0.3	<DL	<DL	<DL	0.3	0.1	0.1	1.3	1.1
BG-7	<DL	0.3	<DL	<DL	<DL	0.2	0.1	0.1	0.6	0.6
BG-8	<DL	0.7	<DL	0.3	<DL	0.6	0.1	0.1	1.2	1.6
BG-9	0.4	0.5	<DL	<DL	<DL	3.3	<DL	<DL	3.8	3.8
BG-10	0.2	0.2	<DL	<DL	<DL	0.7	0.2	0.2	2.6	2.3
BG-11	<DL	0.3	<DL	<DL	<DL	0.3	<DL	<DL	1.5	1.0
BG-12	<DL	0.2	<DL	<DL	<DL	<DL	<DL	<DL	<DL	<DL
BG-13	0.1	0.2	0.1	<DL	0.1	0.3	0.1	0.1	0.5	0.7

Unknowns beside TID	**A-1**	**A-2**	**A-3**	**A-4**	**A-5**	**A-6**	**A-7**	**A-8**	**B-1**	**B-2**
DP-4	7.9	18.3	8.1	5.8	2.9	19.8	0.4	0.6	15.9	21.0
DP-5	7.1	9.7	4.2	3.5	1.4	10.4	0.2	0.2	14.1	20.1
DP-6	<DL	<DL	1.3	0.6	0.3	3.4	<DL	<DL	7.5	7.1

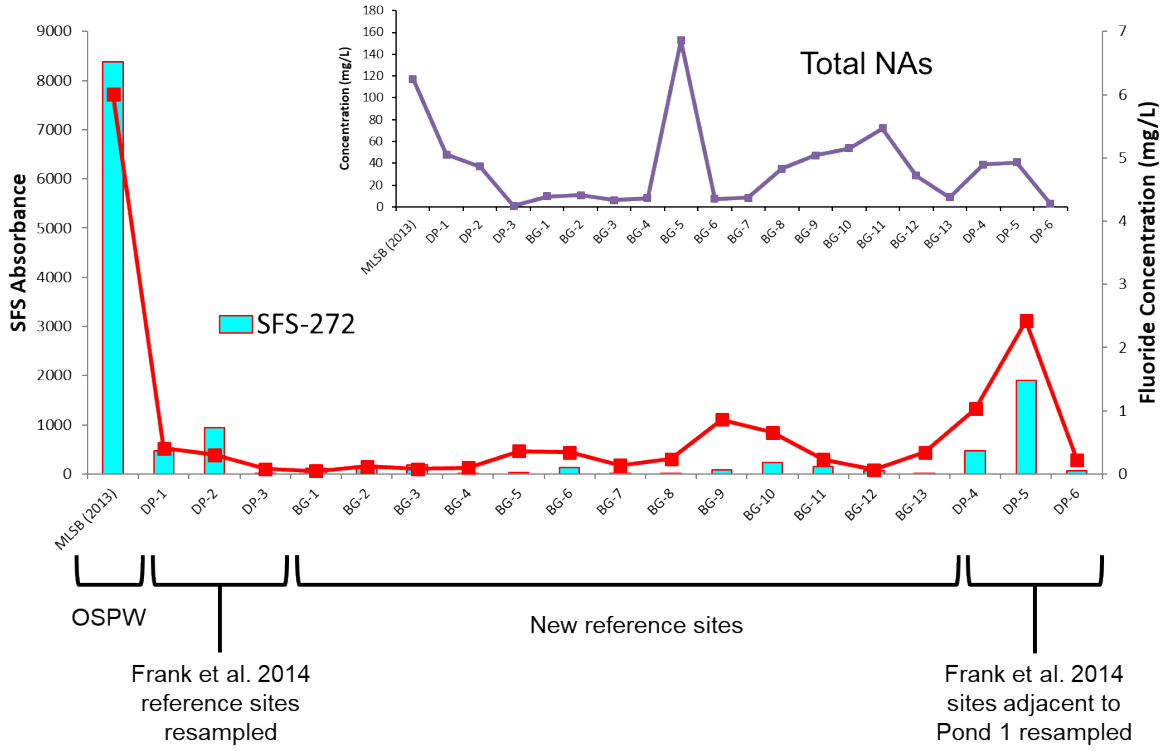

Figure S4. Synchronous fluorescence spectroscopy (SFS) absorbances, Fluoride and total Naphthenic Acid concentrations measured in the sample groups investigated.

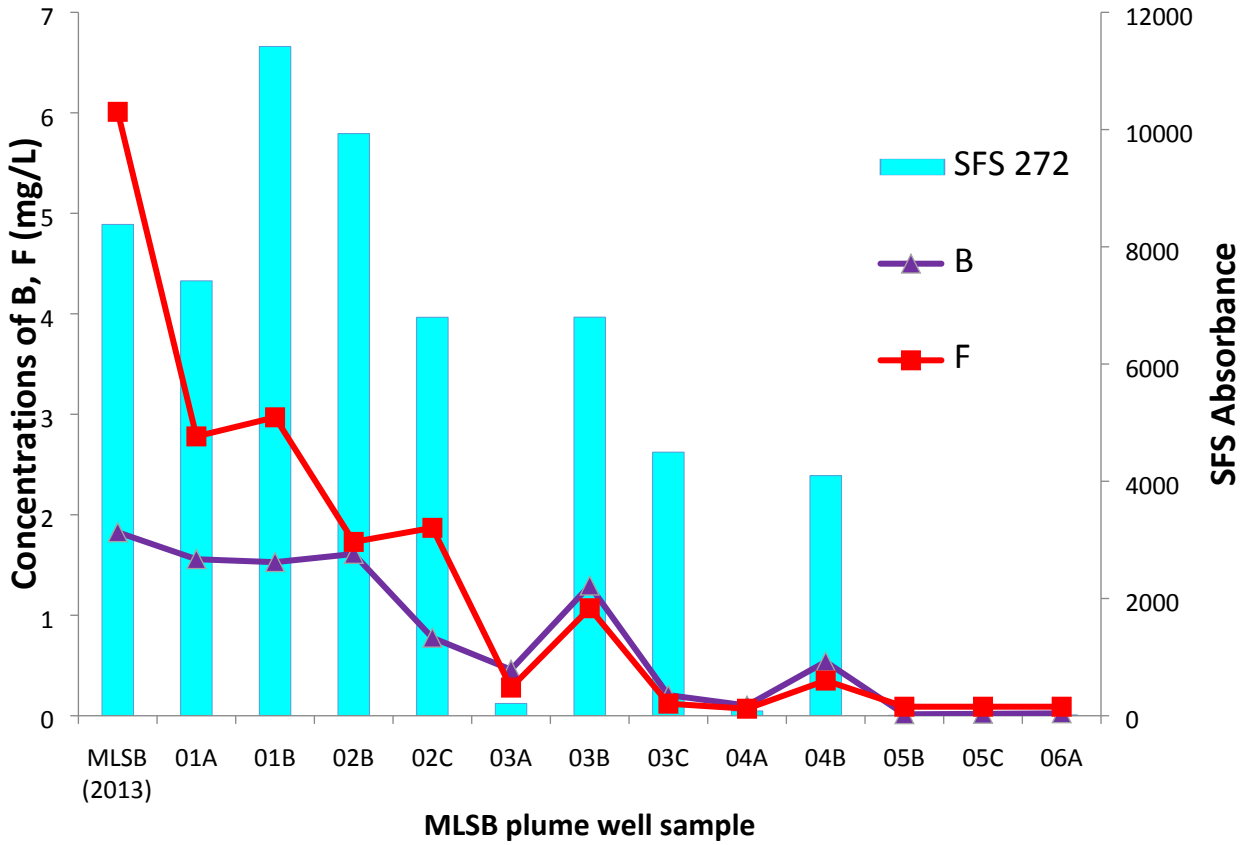

Figure S5. Concentrations of Boron and Fluoride, and SFS absorbances determined for well samples of the MLSB plume monitoring network and the OSPW-source MLSB tailings pond.

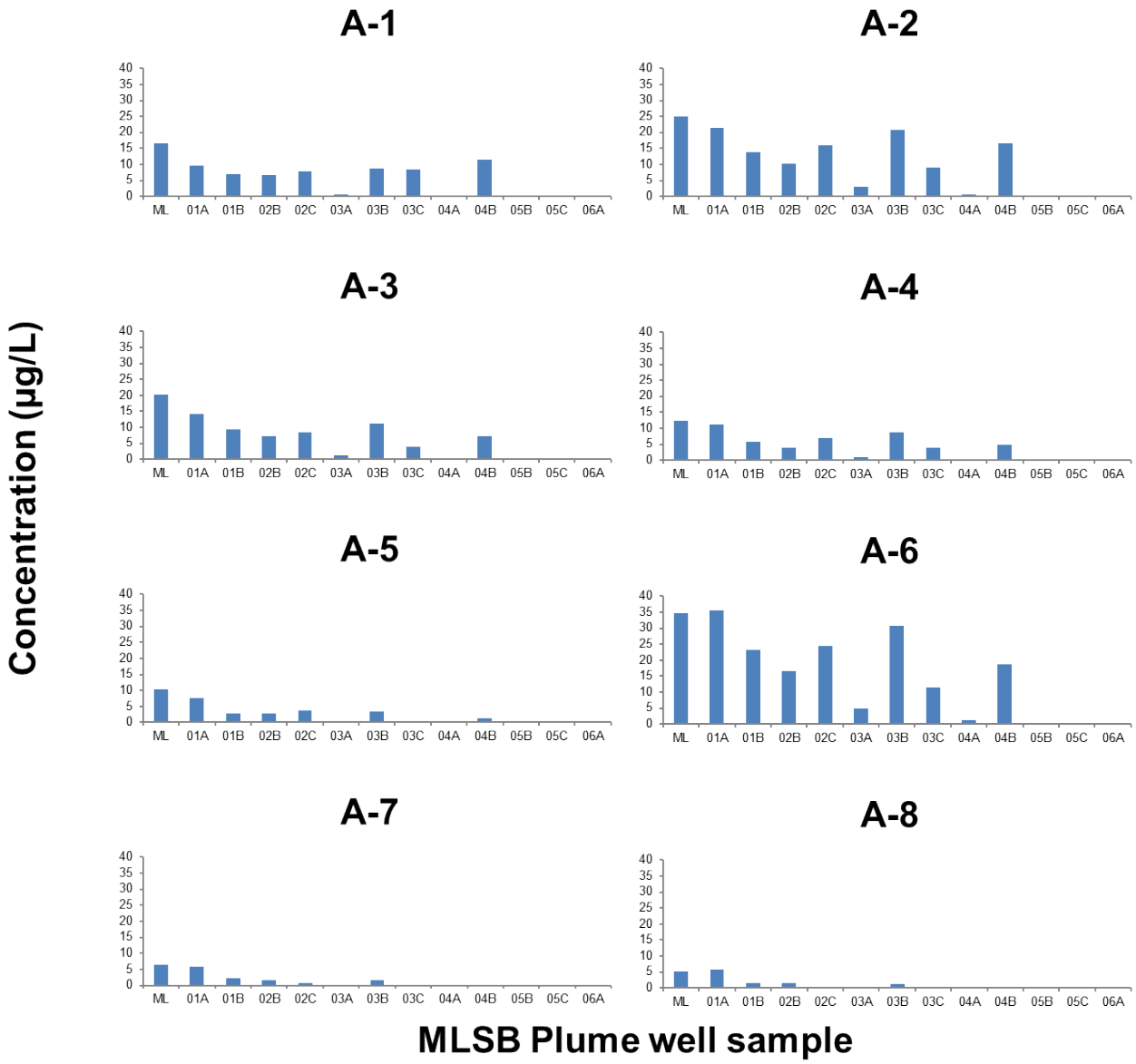

Figure S6. Concentrations of Family A isomers detected in MLSB Plume well samples. "ML" designates the OSPW sample from the source pond, (MLSB 2013), collected at the time of well sampling.

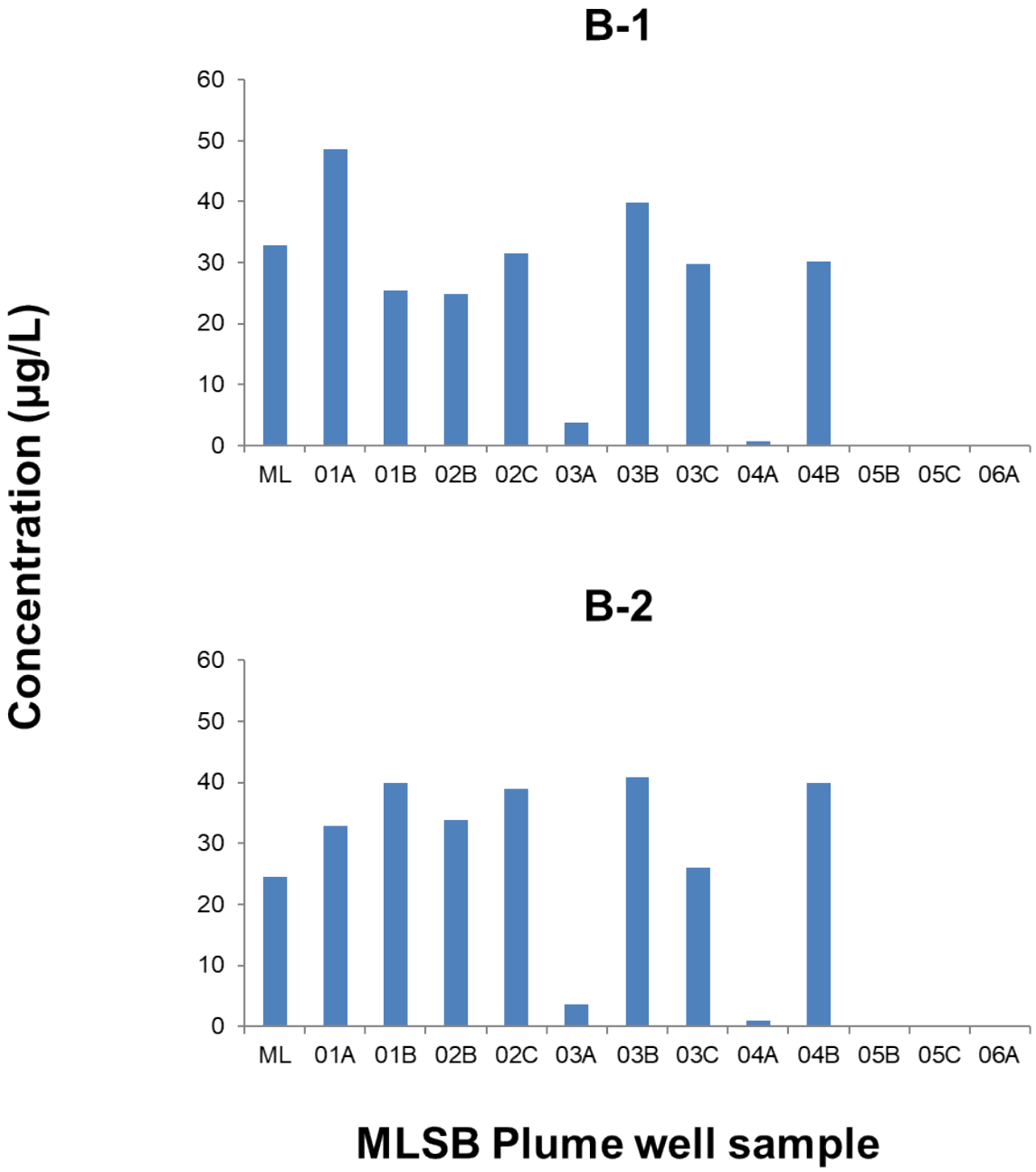

Figure S7. Concentrations of Family B isomers detected in MLSB Plume well samples. "ML" designates the OSPW sample from the source pond, MLSB, collected at the time of well sampling.

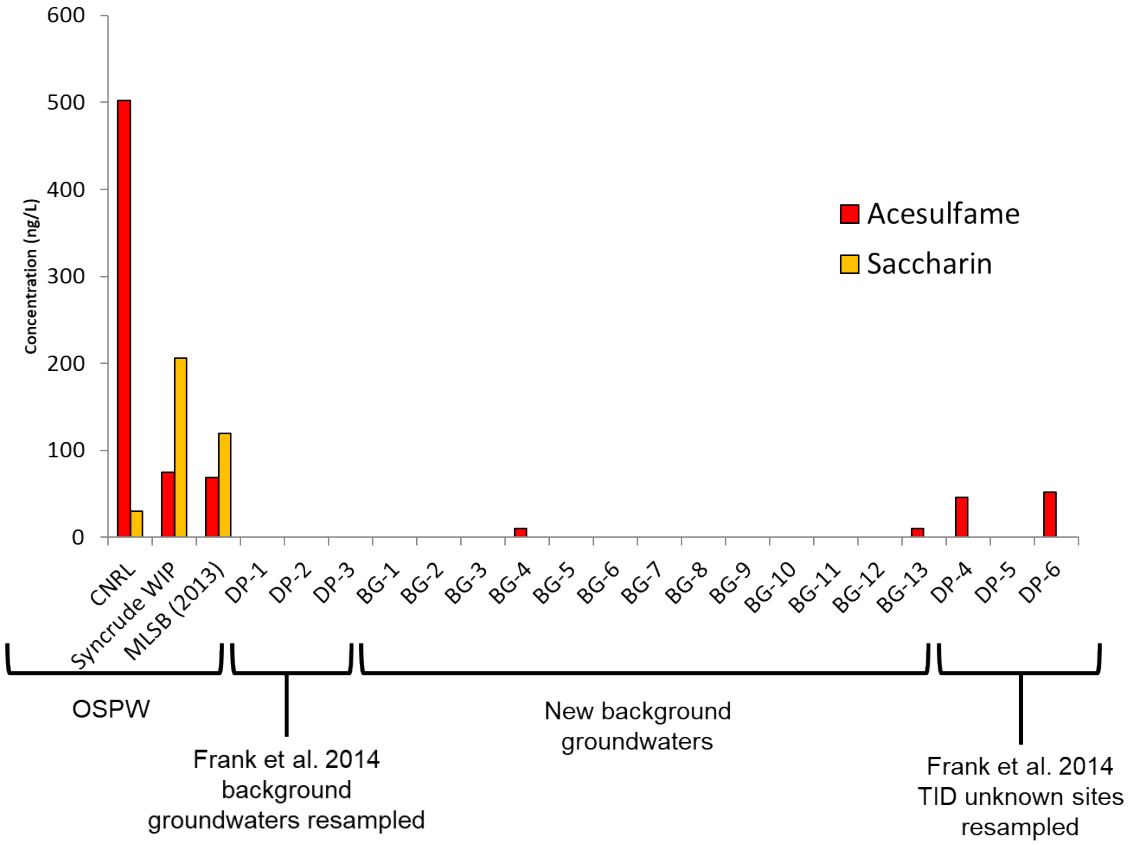

Figure S8. Concentrations of artificial sweeteners in the OSPW, background groundwaters and Unknown groundwater samples beside Tar Island Dyke.

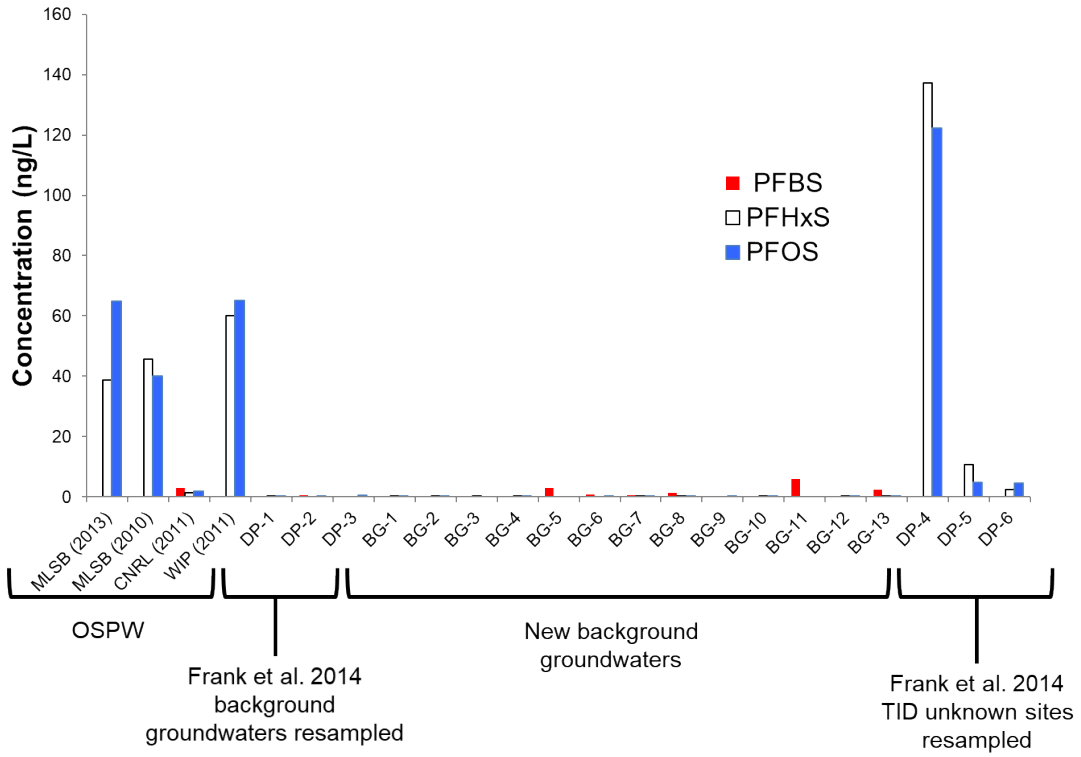

Figure S9. Concentrations of PFAS compound classes in OSPW, background groundwaters and Unknown groundwater samples beside Tar Island Dyke.

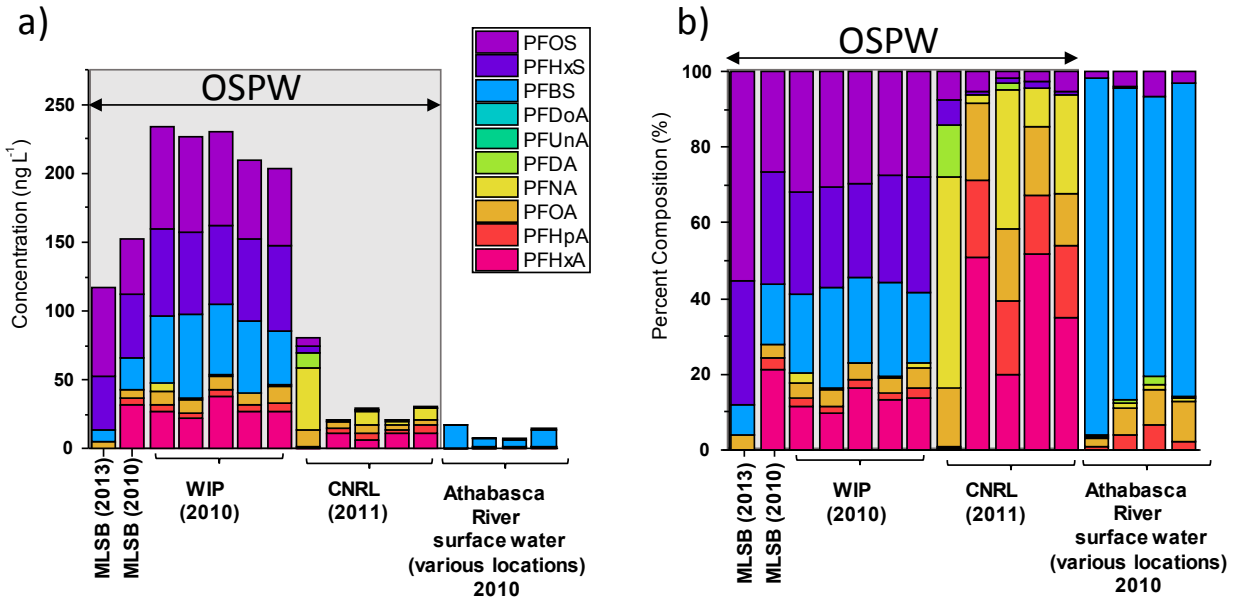

Figure S10. Congener-specific PFAS in Oil Sands Process Water compared to surface waters in the Athabasca River expressed as a) concentration and b) composition. Athabasca River water samples were collected in late March 2010 at four sites ranging from 55° 5' 25.10 "N, 112° 52' 53.80" W to 57° 25' 29.40 "N, 111 °38' 41.20"W. Legend applies to both panels.

Table S7. Pearson correlation matrix for the sums of the Family A and B acids, as methyl esters, expressed relative to the MLSB OSPW sample in the MLSB plume wells. Other metrics included from Figure 4 were as concentration values. Pearson correlations. n=13. r-values are above the diagonal; p-values are below the diagonal.

	MLSB/ Family A	MLSB/ Family B	Chloride	Sodium	Total NA	Acesulfame	Saccharin	Total PFBS	Total PFHxS	Total PFOS
MLSB/Family A	1	0.860	0.840	0.923	0.792	0.221	0.826	0.237	0.811	0.868
MLSB/Family B	<0.001	1	0.712	0.908	0.934	-0.177	0.544	0.393	0.954	0.963
Chloride	<0.001	0.006	1	0.851	0.730	0.299	0.778	0.128	0.771	0.730
Sodium	<0.001	<0.001	<0.001	1	0.935	0.061	0.744	0.196	0.940	0.927
Total NA	0.001	<0.001	<0.001	<0.001	1	-0.210	0.570	0.244	0.985	0.954
Acesulfame	0.468	0.562	0.321	0.844	0.490	1	0.595	-0.201	-0.153	-0.103
Saccharin	<0.001	0.055	0.002	0.004	0.042	0.032	1	0.004	0.594	0.615
Total PFBS	0.436	0.184	0.676	0.522	0.422	0.510	0.990	1	0.264	0.413
Total PFHxS	0.001	<0.001	0.002	<0.001	<0.001	0.617	0.320	0.383	1	0.945
Total PFOS	<0.001	<0.001	0.005	<0.001	<0.001	0.737	0.025	0.161	<0.001	1