

Wastewater Treatment for Nutrients and Pathogens in a Demonstration-Scale Outdoor Constructed Wetland System

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Materials and Methods:

Preparation of reference materials:

Phosphate sorbed on calcite (labeled PSC) reference materials were synthesized by adding 5 g calcite powder to 0.25 L of a 1 mg L⁻¹ PO₄-P solution (for PSC 0.004%), 5 g calcite powder to 1 L of a 250 mg L⁻¹ PO₄-P solution (for PSC 1%), and 2.5 g calcite powder to 1 L of a 1000 mg L⁻¹ PO₄-P solution (for PSC 8%) and shaking (200 rpm) for 48 hours. The white precipitates were collected on a Whatman grade no.4 filter paper and freeze dried before characterization by Fourier transform infrared spectroscopy (FTIR) and field emission scanning electron microscopy (SEM; Leo 1530) with energy dispersive X-ray (EDX). The quantity of phosphate in the PSC was estimated from the initial and final concentrations of P in the solution.

Solid-phase analyses:

The spent BOFS samples and the reference materials were examined using SEM-EDX FTIR. Small quantities of the freeze-dried samples were mounted on double-sided carbon tape, which was pasted on SEM stubs. These samples were gold plated before analysis. SEM images were collected in back-scattered electron (BSE) mode. Three to five images and corresponding EDX spectra were collected for each sample. Transparent 1 cm diameter KBr pellets were prepared using a 13 mm pellet die by pressing a mixture of the ground samples and KBr (sample to KBr ratio was approximately 1:100) with 5 to 6 tonnes pressure for 1 min. A Bruker Tensor 27 infrared spectrometer was used to examine the KBr pellets. The data acquisition parameters for these analyses included 4 cm⁻¹ resolution, 16 scans, and wave numbers between 4000 and 400 cm⁻¹. The spectrum for blank KBr was subtracted from each spectrum to minimize the influence of the KBr.

XANES analysis was conducted at the Canadian Light Source, Saskatoon, Canada. The energy of the storage ring in this facility was 2.9 GeV. Soft X-ray micro-characterization beamline (SXRMB; 06B1-1), covering the energy region of 1700-10000 eV with photon resolution of 0.2 eV and a beam spot size of 300 µm x 300 µm, was used to record XANES data for P and Ca. Normally the edge energy of Na₄P₂O₇ is set to 2152.4

eV and the other PO₄ spectra are measured ~2152 eV in the SXRMB beamline at the Canadian Light Source. The edge energies of the reference materials and sample spectra were within 0.5 eV of the PO₄ edge (2152 eV). All samples were freeze-dried and outer layers of the spent media were subsampled. Both samples and reference materials were ground to ~50 µm and spread on carbon double sided tape previously pasted on a sample holder. The scan parameters used for P-XANES included: energy range, 2114-2205 eV; edge energy, 2145.5 eV; step size, 2114-2142 eV @ 1 eV, 2142-2170 eV @ 0.1 eV, and 2170-2205 eV @ 0.5 eV; counting times, 1-4 s; number of scans, 2-3. While the scan parameters used for Ca-XANES included: energy range, 3981-4190 eV; edge energy, 4038.5 eV; step size, 3980.31-4034.31 eV @ 2 eV, 4034.31 -4090.31 eV @ 0.2 eV, and 4090.31-4190.31 eV @ 1 eV; counting times, 1 -4 s; number of scans, 2-3. XANES data were collected in Total Electron Yield (TEY) and Fluorescent Yield (FY) mode. A monochromator with Si (111) crystals and a flux of >1x10¹¹ photons s⁻¹ was used to collect spectra from the experimental samples and the standards. To improve signal-to-noise ratios the spectral data were averaged over 2-3 scans. The reference materials used for the FTIR analysis also were used for the XANES experiments. The XANES spectra of the unknown samples were compared to spectra for the reference materials by linear combination fitting using the Athena software package, version 0.8.56 [55].

Results:

FTIR results:

In the FTIR spectra obtained from the spent media collected from the columns from year 1, carbonate bands at 712-713 cm⁻¹, 1424-1428 cm⁻¹ and 874-875 cm⁻¹, and carbonate overtones or combination bands at 1796-1799 cm⁻¹, 2513-2517 cm⁻¹, 2871-2876 cm⁻¹ and 2979-2986 cm⁻¹ were observed, providing a strong indication of the presence of calcite. However, a shoulder of the carbonate ν_2 band at 858 cm⁻¹ (a unique feature of aragonite [80]; in sample C2-S10 (Column 2-Sample 10), in addition to the sharp ν_2 band at 875 cm⁻¹ and ν_4 band at 712 cm⁻¹, suggested that this sample may also contain a small quantity of aragonite.

Water Quality Guidelines/Objectives:

The guideline value for cBOD₅ is 25 mg L⁻¹, based upon performance data collected in 1983 of sewage treatment works in operation in Ontario [103]. The effluent design objectives for TP was 1 mg L⁻¹ (for extended aeration with TP removal) and no objective was set for NH₃-N based on the expected effluent quality under optimum conditions. Our treatment system has an aeration option before the BOFS cell, however, it is not considered as extended aeration.

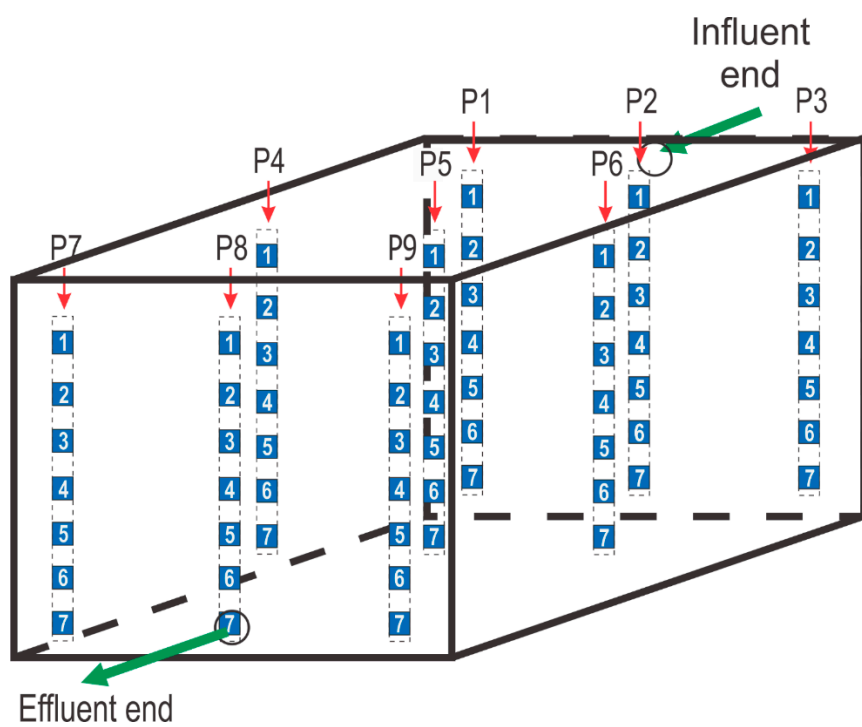


Figure S1. Locations of solid phase spent media collected from the BOFS cell.

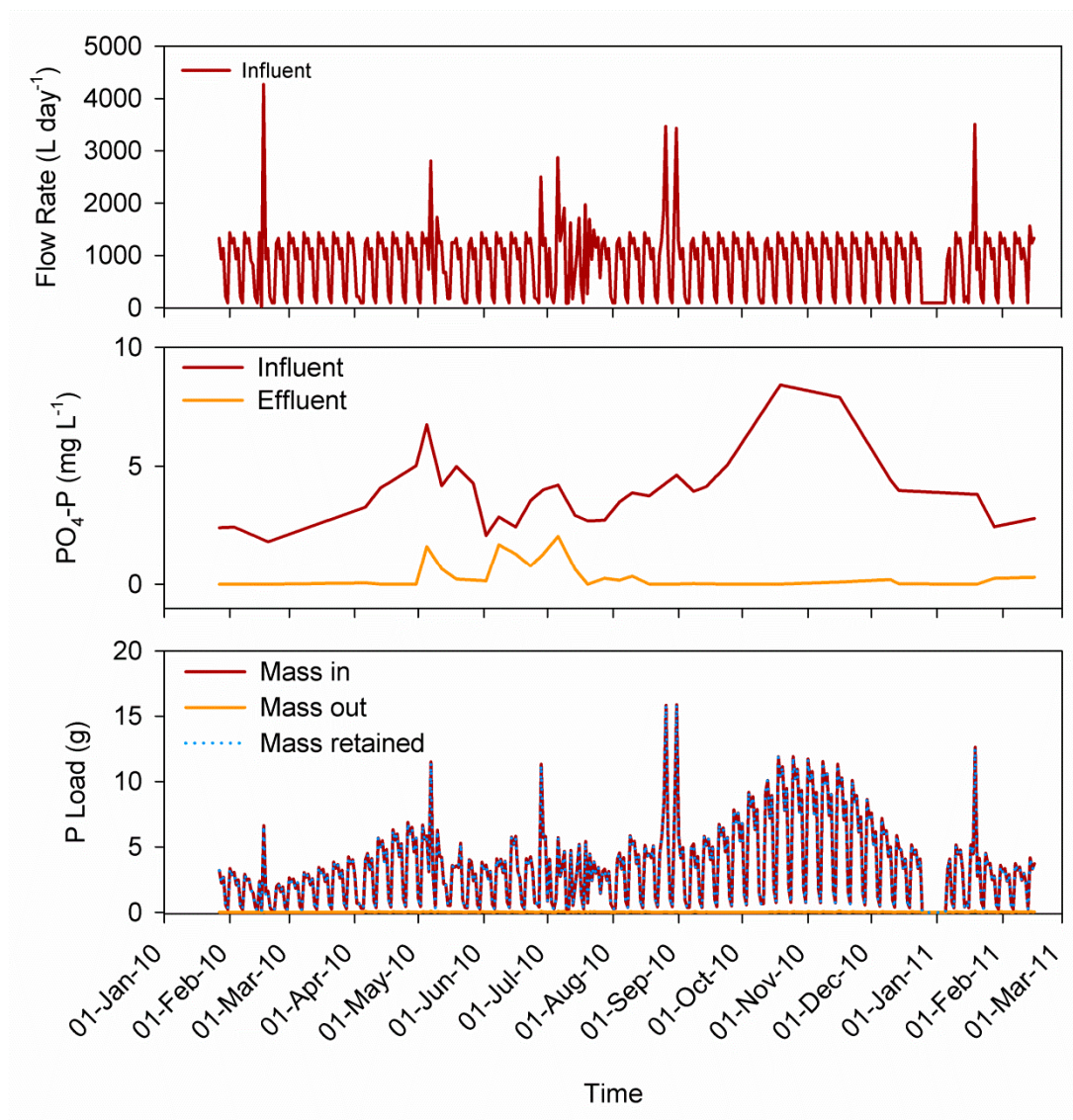


Figure S2. Flow rate, PO₄-P concentrations in the influent and effluent, and P load in BOFS Cell. .

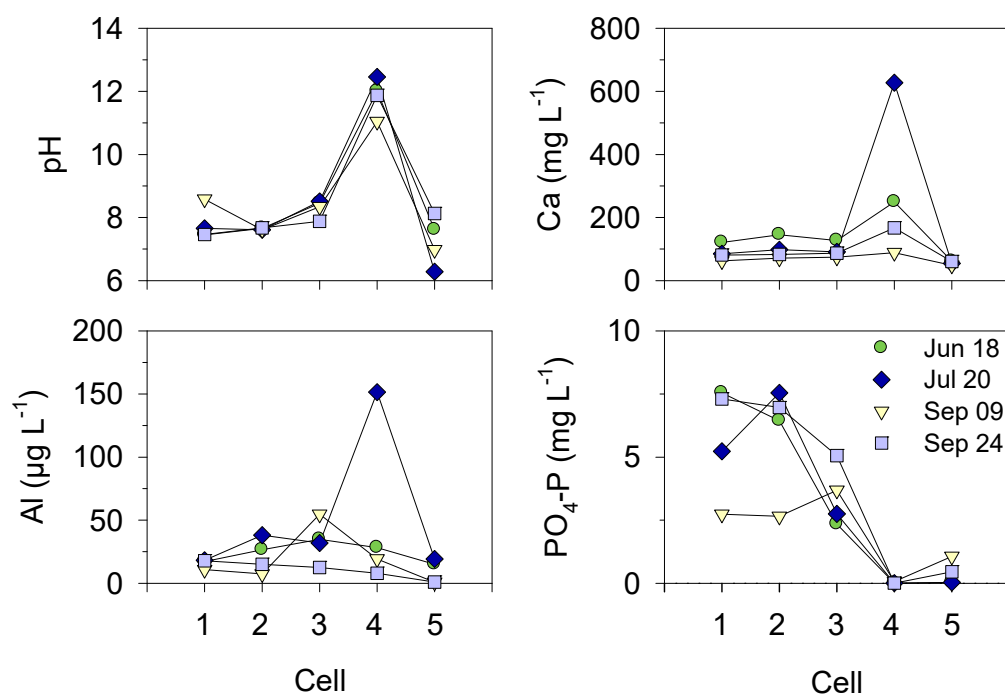


Figure S3. pH values, Ca, Al, and PO₄-P concentrations versus distance (Cells 1-5) along the flow path.

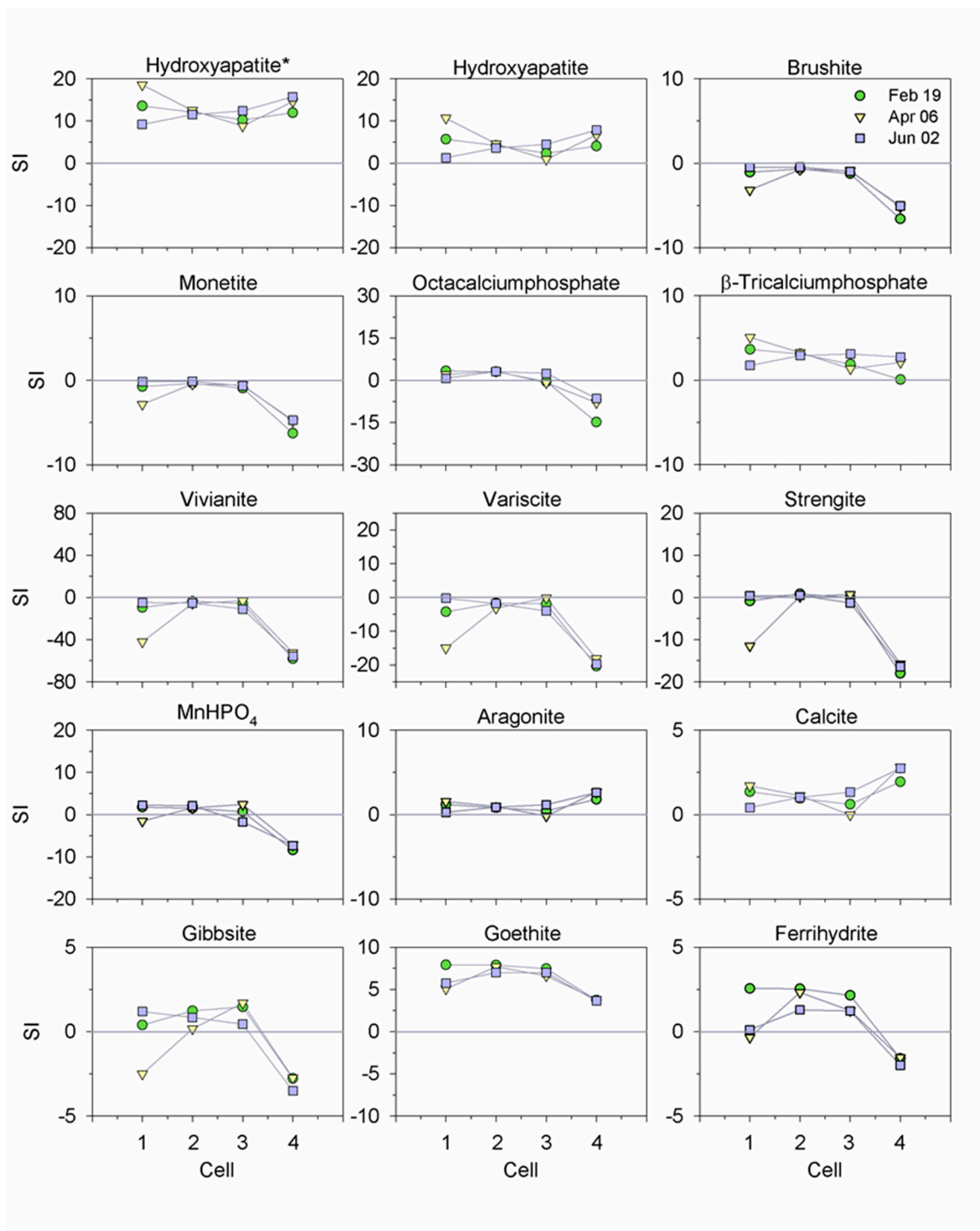


Figure S4. Saturation indices for calcium phosphate and other related phases (calculated using PHREEQCI) along the treatment cells during three sampling events. Saturation index of hydroxyapatite (indicated by *) was plotted using a modified $\log K_{sp}$ value [31]. .

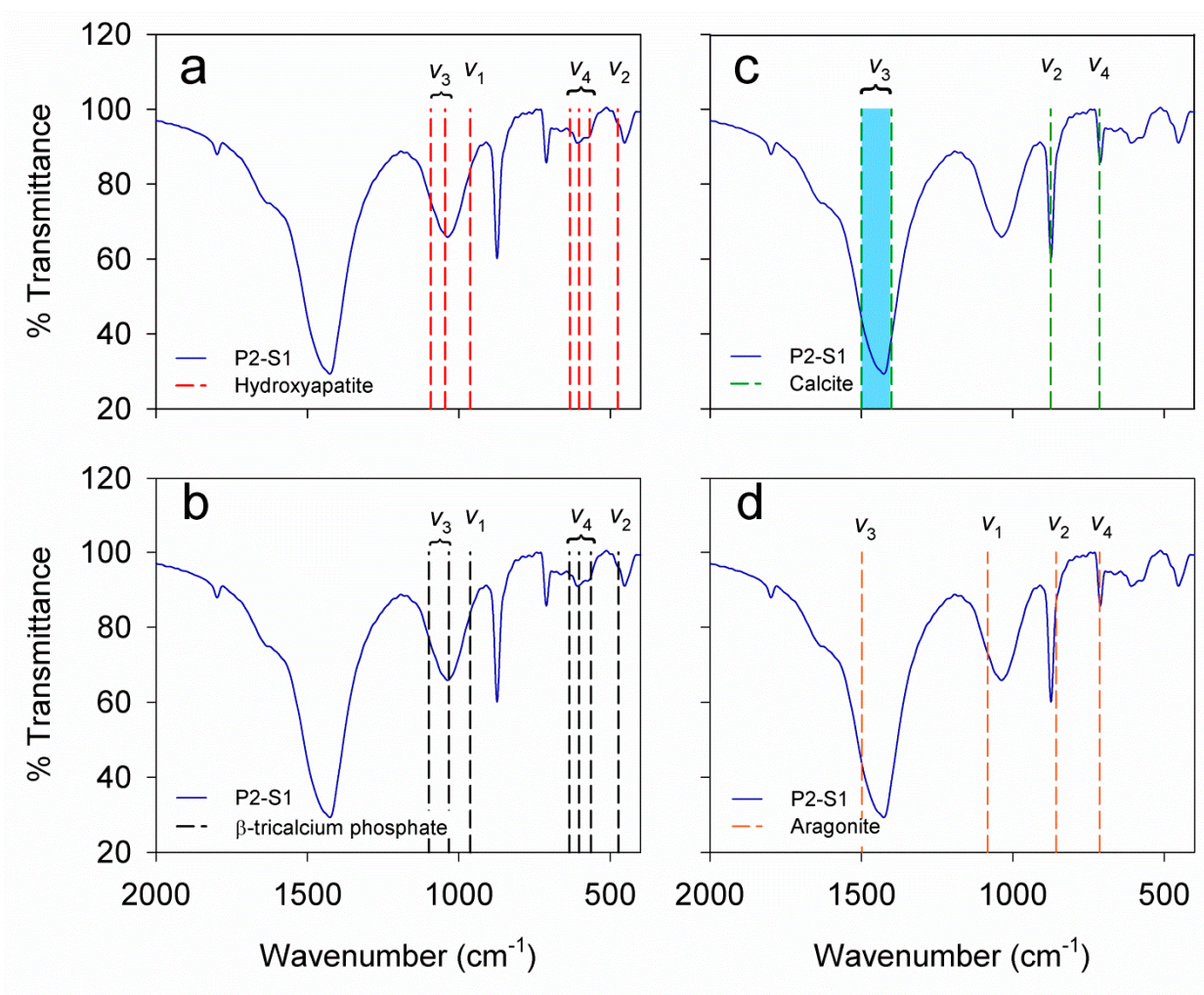


Figure S5. FTIR spectrum of sample with highest mass and reference materials and sample P2-S1, with phosphate vibrational bands for a) HAP-S, b) β -TCP, c) calcite, d) aragonite. Vertical dotted lines in "a" and "b" represent phosphate vibrational bands, while vertical dotted lines in "c" and "d" represent carbonate vibrational bands.

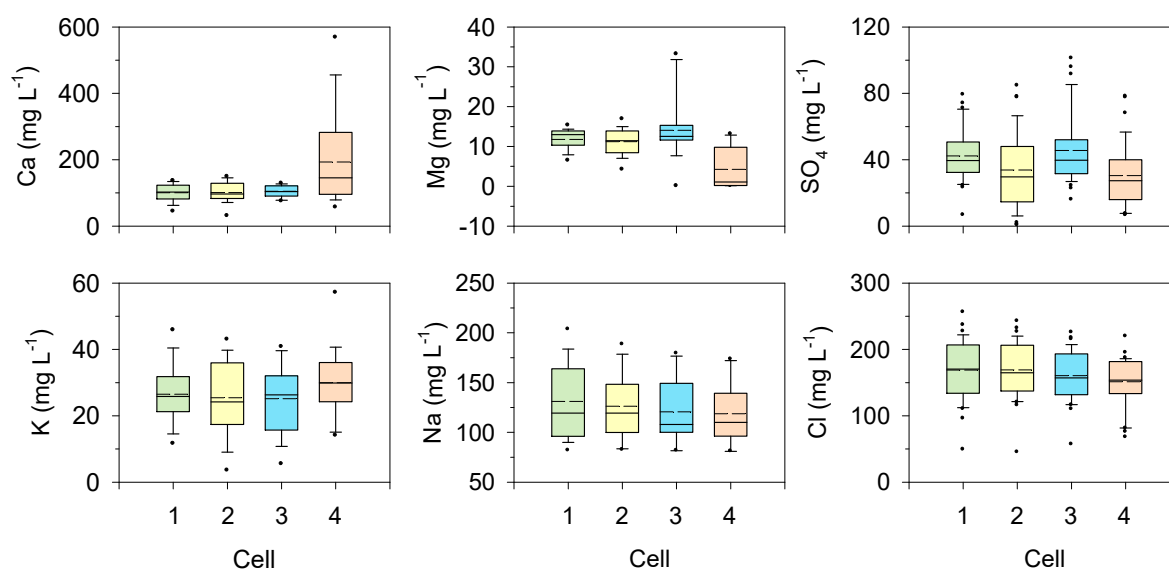


Figure S6. Box plots of Ca, Na, Mg, K, SO₄, and Cl concentrations versus distance (Cells 1-4) along the flow path. Boxes represent 50% of the data (between the first and third quartiles). Horizontal solid lines and broken lines on the boxes represent median and mean concentrations. Bars extend from the box to the highest/lowest value within 1.5 * inter-quartile range (IQR). Points (outliers) are values > 1.5 * IQR. Top- and bottom-most dots represent maximum observation above upper fence and minimum observation below lower fence. .

Table S1. K_{sp} values of the calcium phosphate minerals added to the PHREQCI database. .

Mineral Phases	Reactions	$\log K_{sp}$	References
Hydroxyapatite*	$\text{Ca}_5(\text{PO}_4)_3\text{OH} + \text{H}^+ = 5\text{Ca}^{2+} + 3\text{PO}_4^{3-} + \text{H}_2\text{O}$	-36.31	[31]
Brushite	$\text{CaHPO}_4 \cdot 2\text{H}_2\text{O} = \text{Ca}^{2+} + \text{PO}_4^{3-} + 2\text{H}_2\text{O} + \text{H}^+$	-18.91	
Monetite	$\text{CaHPO}_4 = \text{Ca}^{2+} + \text{PO}_4^{3-} + \text{H}^+$	-19.24	
Octacalciumphosphate	$\text{Ca}_8\text{H}_2(\text{PO}_4)_6 \cdot 5\text{H}_2\text{O} = 8\text{Ca}^{2+} + 5\text{H}_2\text{O} + 6\text{PO}_4^{3-} + 2\text{H}^+$	-93.95	
β -tricalciumphosphate	$\text{Ca}_3(\text{PO}_4)_2 = 3\text{Ca}^{2+} + 2\text{PO}_4^{3-}$	-28.94	
Variscite	$\text{AlPO}_4 \cdot 2\text{H}_2\text{O} = \text{Al}^{3+} + \text{PO}_4^{3-} + 2\text{H}_2\text{O}$	-21	[67]

Table S2. Physical and chemical properties of the BOFS materials used in the experiments [54].

BOFS	
Source: Stelco Hamilton	
Physical properties	
Particle size (mm)	Surface area (m ² g ⁻¹)
4-2	6.05
2-1	8.98
1-0.5	11.5
<0.5	13.9
Particle size (mm)	Mass weighted surface area (m ² g ⁻¹)
4-2	154
2-1	196
1-0.5	250
<0.5	429
Density (g cm ⁻³):	3.49
Chemical properties	
Chemical composition	wt. %
CaO	33.8
Fe ₂ O ₃	24.3
SiO ₂	11.3
MgO	9.62
MnO	4.08
Al ₂ O ₃	7.43
P ₂ O ₅	-
TiO ₂	0.38
K ₂ O	0.03
Na ₂ O	0.07
Cr ₂ O ₃	-
Loss on ignition	8.96

Table S3. Physical and chemical properties of the ZVI materials used in the experiments [54].

ZVI	
Source: Connelly GPM Inc., Chicago, Illinois, USA	
Physical properties	
Particle size (mm)	Surface area (m ² g ⁻¹)
0.297-2.38	4.4
Density (g cm ⁻³):	3.49
Chemical properties	
Chemical composition	wt. %
Iron/Iron Oxide	Balance
Total Carbon	2.48
Manganese	0.93
Sulphur	0.12
Phosphorus	ND
Silicon	0.35
Nickel	>0.01
Chromium	>0.01
Vanadium	ND
Molybdenum	0.33
Copper	0.10
Aluminum	>0.01
Magnesium	0.01
Boron	0.01
Zinc	0.01
Zirconium	0.01
ND= Not Detectable	

Table S4. Mean (arithmetic) concentrations, concentration ranges (expressed as average of maximum and minimum values), standard deviation, confidence Norm, and 95% confidence interval of pH, alkalinity, conductivity, and other target parameters including PO₄, NH₃-N, NO₃-N, NO₂-N, Cl, and SO₄ in the effluents of Cells 1, 2, 3, and 4.

Unit		Effluents				
		Cell 1	Cell 2	Cell 3	Cell 4	
pH		Mean	7.61	7.81	7.94	11.4
		Range	7.89±1.06	8.42±1.20	7.65±0.94	10.88±1.47
		St.Dev.	0.51	0.41	0.40	0.77
		Confidence.Norm	0.16	0.12	0.12	0.24
		Confidence interval	7.61±0.16	7.81±0.12	7.94±0.12	11.40±0.24
Alk	mg L ⁻¹ as CaCO ₃	Mean	550	505	237	738
		Range	661±389	494±246	284±256	1225±1135
		St.Dev.	150	123	99	551
		Confidence.Norm	46	38	31	171
		Confidence interval	550±46	505±38	239±31	654±171
Conductivity	µS cm ⁻¹	Mean	1636	1546	1333	4052
		Range	1669±665	1412±610	1268±449	4661±3500
		St.Dev.	322	287	248	2270
		Confidence.Norm	100	89	77	703
		Confidence interval	1636±100	1546±89	1333±77	4052±703
PO ₄ -P	mg L ⁻¹	Mean	6.32	6.74	3.86	0.33
		Range	12.9±9.32	8.40±6.21	5.12±3.29	1.03±1.03
		St.Dev.	3.18	2.27	1.38	0.06
		Confidence.Norm	0.92813	0.66319	0.40445	0.01809
		Confidence interval	6.32±0.93	6.09±0.66	3.65±0.40	0.04±0.018
TP	mg L ⁻¹	Mean	8.41	7.23	3.64	1.13
		Range	13.5±9.83	10.2±8.16	4.98±3.01	3.67±3.64
		St.Dev.	2.34	3.01	1.41	1.56
		Confidence.Norm	0.9	1.1	0.5	0.6
		Confidence interval	7.3±0.9	6.8±1.1	3.6±0.5	1.3±0.6
NH ₃ -N	mg L ⁻¹	Mean	47.9	38.3	3.64	8.29
		Range	62.2±47.2	54.7±46.9	15.0±15.0	29.4±29.2
		St.Dev.	26	19	7.5	14
		Confidence.Norm	8.0	6.1	2.3	4.3
		Confidence interval	48±8.0	38±6.1	3.6±2.3	8.3±4.3
NO ₃ -N	mg L ⁻¹	Mean	3.34	2.12	41.52	34.60
		Range	22.6±22.6	18.7±18.7	53.9±39.0	41.6±38.7
		St.Dev.	8.3	6.7	18	16
		Confidence.Norm	2.4	2.0	5.3	4.7
		Confidence interval	3.3±2.4	2.2±2.0	42±5.3	35±4.7
NO ₂ -N	mg L ⁻¹	Mean	0.83	4.53	2.07	2.62
		Range	8.59±8.58	36.7±36.7	12.0±12.0	3.48±3.24
		St.Dev.	2.7	16	5.9	2.0
		Confidence.Norm	0.8	4.9	1.8	0.6
		Confidence interval	0.8±2.4	3.3±4.2	2.1±1.9	2.7±0.6
Cl	mg L ⁻¹	Mean	169	169	161	151
		Range	152±104	144±98.6	141±84.3	144±76.0
		St.Dev.	45	41	38	34
		Confidence.Norm	14	13	12	11
		Confidence interval	169±14	169±13	161±12	151±11
SO ₄	mg L ⁻¹	Mean	42.3	33.9	45.6	30.5
		Range	42.8±36.3	42.6±42.0	58.5±42.6	42.3±35.8
		St.Dev.	15	22	20	19
		Confidence.Norm	5	7	6	6
		Confidence interval	42±5	34±7	46±6	30±6

Table S5. Vibrational bands of carbonate and phosphate and the corresponding wavenumbers in the FTIR spectra of the surface materials.

Carbonate	P1-S1	P2-S1	P 3-S1	P4-S1	P5-S1	P6-S1	P7-S1	P8-S1	P9-S1
ν_4	712	712	712	712	712	712	712	713	712
ν_2	873	874	873	873	873	872	873	874	873
ν_3	1426	1426	1425	1423	1426	1422	1425	1425	1426
$\nu_1+\nu_4$	1798	1797	1800	1799	1798	1799	1798	1798	1799
$\nu_1+\nu_3$ or $2\nu_2+\nu_4$	2514	2514	2515	2514	2515	2514	2514	2515	2515
$2\nu_3$	2874	2874	2875	2875	2874	2876	2875	2874	2875
$2\nu_3$	2980	2980	2980	2982	2981	2982	2978	2982	2980, 2924
Phosphate									
ν_3	1035	1037	1035	1037	1035	1037	1039	1034	1030
ν_1		-							
ν_4	602, 583, 567	632, 608, 574	604, 584, 572	604, 576, 568	635, 605, 588, 573	604, 578, 569	606, 580, 568	605, 597, 587, 579, 569	632, 604, 595, 587, 568
ν_2	450	475, 453	451	450	451	450	461	454	459

Table S6. Mass change of PO₄-P observed in different components of the treatment system. The +ve values indicate mass losses from the system component.

	PO ₄ -P mass in (mg)	PO ₄ -P mass out (mg)	PO ₄ -P mass change (mg)	PO ₄ -P mass change (Kg)	PO ₄ -P mass % change
Overall treatment system	2003663	89904	1913759	1.91	95.51
Cell 2	2003663	1978166	25497	0.03	1.27
Cell 3	1978166	1450086	528080	0.53	26.70
Cell 4	1450086	89904	1360182	1.36	93.80

Table S7. Linear combination fitting results of P-XANES for spent BOFS samples.

Linear combination fits to P3-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, CPD	3.57 x10 ⁻³	0.38	-	-	-	0.988	0.012	-
PSC 1%, CPDD, β -TCP	3.5757x10 ⁻³	0.38	-	0	0.003	0.997	-	-
CPD, CPDD, β -TCP	1.1257x10 ⁻²	1.20	-	0.727	0.259	-	0.014	-
CPD, HAP, α -TCP, β -TCP	1.20 x10 ⁻²	1.29	0.039	0.733	-	-	0.228	0
Linear combination fits to P4-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
α -TCP, PSC 1%	6.37 x10 ⁻³	0.67	0.185	-	-	0.815	-	-
CPD, α -TCP, CPDD, PSC 1%,	6.66 x10 ⁻³	0.70	0.172	-	0.060	0.768	0	-
CPD, α -TCP, PSC 1%, β -TCP	7.91 x10 ⁻³	0.83	0.229	0.190	-	0.581	0	-
HAP, α -TCP, PSC 1%, β -TCP	8.10 x10 ⁻³	0.85	-	0.207	-	0.555	0.239	0
Linear combination fits to P6-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, β -TCP	3.08 x10 ⁻³	0.33	-	0.197	-	0.803	-	-
α -TCP, CPDD, β -TCP	6.82 x10 ⁻³	0.73	0.046	0.884	0.070	-	-	-
CPDD, β -TCP	6.88 x10 ⁻³	0.74	-	0.897	0.103	-	-	-
CPD, β -TCP	7.23 x10 ⁻³	0.78	-	0.96	-	-	0.04	-
Linear combination fits to P8-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
HAP, PSC 1%, β -TCP	1.55 x10 ⁻²	1.64	-	0.497	-	0.393	-	0.110
PSC 1%, β -TCP	1.56 x10 ⁻²	1.65	-	0.652	-	0.348	-	-
HAP, PSC 1%	1.62 x10 ⁻²	1.71	-	-	-	0.694	-	0.306
CPD, CPDD, PSC 1%, β -TCP	1.62 x10 ⁻²	1.72	-	0.678	0	0.221	0.101	-

Notes: HAP- Hydroxyapatite; PSC 1% - phosphate sorbed on calcite 1%; α -TCP - alpha-tricalcium phosphate; β -TCP - beta-tricalcium phosphate; CPD - calcium phosphate dibasic CPDD - calcium phosphate dibasic dihydrate.

Table S8. Linear combination fitting results of Ca-XANES for spent BOFS samples.

Linear combination fits to P3-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, α -TCP	1.73 x10 ⁻³	0.068	0.144	-	-	0.856	-	-
PSC 1%, CPD	2.56 x10 ⁻³	0.101	-	-	-	0.975	0.025	-
PSC 1%, CPDD	2.60 x10 ⁻³	0.103	-	-	0.010	0.990	-	-
PSC 1%, CPD, CPDD, HAP	2.60 x10 ⁻³	0.103	-	-	0.014	0.985	0.001	-
Linear combination fits to P4-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, CPDD	1.18 x10 ⁻³	0.05	-	-	0.062	0.938	-	-
PSC 1%, β -TCP, CPDD, HAP	1.21 x10 ⁻³	0.053	-	0.060	0.009	0.931	-	0
PSC 1%, HAP	1.37 x10 ⁻³	0.060	-	-	-	0.945	-	0.055
PSC 1%, CPD	1.82 x10 ⁻³	0.080	-	-	-	0.933	0.067	-
Linear combination fits to P6-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, CPDD	7.96 x10 ⁻⁴	0.035	-	-	0.090	0.910	-	-
PSC 1%, CPD, β -TCP, α -TCP	9.76 x10 ⁻⁴	0.043	0.012	0.089	-	0.894	0.006	-
PSC 1%, α -TCP, β -TCP	9.77 x10 ⁻⁴	0.043	0.016	0.091	-	0.893	-	-
PSC 1%, β -TCP, CPD	9.78 x10 ⁻⁴	0.043	-	0.087	-	0.899	0.013	-
Linear combination fits to P8-S1								
Combination	R-factor	χ^2	α -TCP (weight)	β -TCP (weight)	CPDD (weight)	PSC 1% (weight)	CPD (weight)	HAP (weight)
PSC 1%, β -TCP, CPDD	1.11 x10 ⁻³	0.049	-	0.021	0.079	0.900	-	-
PSC 1%, CPDD	1.11 x10 ⁻³	0.049	-	-	0.098	0.902	-	-
PSC 1%, β -TCP	1.17 x10 ⁻³	0.052	-	0.107	-	0.893	-	-
PSC 1%, HAP	1.34 x10 ⁻³	0.059	-	-	-	0.908	-	0.092

Notes: HAP- Hydroxyapatite; PSC 1% - phosphate sorbed on calcite 1%; α -TCP - alpha-tricalcium phosphate; β -TCP - beta-tricalcium phosphate; CPD - calcium phosphate dibasic CPDD - calcium phosphate dibasic dihydrate.

Table S9. Mass change of N_T observed in different components of the treatment system. The +ve values indicate mass losses from the system component and the -ve values indicate mass gains within the system component.

	N _T mass in (mg)	N _T mass out (mg)	N _T mass change (mg)	N _T mass change (Kg)	N _T mass % change
Overall treatment system	17921722	17550343	371379	0.37	2.07
Cell 2	17921722	17022165	899557	0.90	5.02
Cell 3	17022165	18591246	-1569081	-1.57	-9.22
Cell 4	18591246	17550343	1040904	1.04	5.60

Table S10. Mean (arithmetic) concentrations, concentration ranges (expressed as average of maximum and minimum values), standard deviation, confidence Norm, and 95% confidence interval of DO, cBOD₅, COD, total coliform, and E. coli in the effluents of Cells 1, 2, 3, and 4.

	Unit		Effluents			
			Cell 1	Cell 2	Cell 3	Cell 4
DO	mg L ⁻¹	Mean	1.53	2.30	8.89	4.64
		Range	3.61±3.48	2.41±2.23	8.59±3.91	5.74±3.54
		St.Dev.	1.8	1.2	1.5	1.9
		Confidence.Norm	0.6	0.4	0.5	0.6
		Confidence interval	1.5±0.6	2.3±0.4	8.9±0.5	4.6±0.7
cBOD ₅	mg L ⁻¹	Mean	62.71	26.4	7.61	0.82
		Range	103±98.5	48.1±46.2	18.9±18.2	1.34±0.84
		St.Dev.	51	28	9	0.4
		Confidence.Norm	17	9	3	0.1
		Confidence interval	63±17	26±9	8±3	0.8±0.1
COD	mg L ⁻¹	Mean	143	72.1	28.4	34
		Range	194±154	154±119	39.2±26.0	64.2±47.8
		St.Dev.	84	54	13	19
		Confidence.Norm	29	18	4	7
		Confidence interval	143±29	72±18	28±4	34±7
Total coliform	CFU 100 mL ⁻¹	Mean	2.00 x10 ⁻⁶	9.04 x10 ⁻⁴	1.71 x10 ⁻³	8.49 x10 ⁻¹
		Range	6.06 x10 ⁻⁶ ±6.06 x10 ⁻⁶	5.87 x10 ⁻⁵ ±5.87 x10 ⁻⁵	9.39 x10 ⁻³ ±9.36 x10 ⁻³	1.21 x10 ⁻³ ±1.21 x10 ⁻³
		St.Dev.	3.05 x10 ⁻⁶	2.16 x10 ⁻⁵	3.58 x10 ⁻³	4.34 x10 ⁻²
		Confidence.Norm	1.04 x10 ⁻⁶	7.35x10 ⁻⁴	1.22 x10 ⁻³	1.48 x10 ⁻²
		Confidence interval	2.00 x10 ⁻⁶ ±1.04 x10 ⁻⁶	9.04 x10 ⁻⁴ ±7.35 x10 ⁻⁴	1.71 x10 ⁻³ ±1.22 x10 ⁻³	8.50 x10 ⁻¹ ±1.48 x10 ⁻²
<i>E. coli</i>	CFU 100 mL ⁻¹	Mean	3.31 x10 ⁻⁵	3.00 x10 ⁻⁴	1.23 x10 ⁻³	1.01 x10 ⁻²
		Range	1.24 x10 ⁻⁶ ±1.24 x10 ⁻⁶	2.42 x10 ⁻⁵ ±2.42 x10 ⁻⁵	1.21 x10 ⁻⁴ ±1.21 x10 ⁻⁴	1.21 x10 ⁻³ ±1.21 x10 ⁻³
		St.Dev.	5.94 x10 ⁻⁵	8.64 x10 ⁻⁴	4.33 x10 ⁻³	4.33 x10 ⁻²
		Confidence.Norm	2.03 x10 ⁻⁵	2.95 x10 ⁻⁴	1.48 x10 ⁻³	1.48 x10 ⁻²
		Confidence interval	3.31 x10 ⁻⁵ ±2.03 x10 ⁻⁵	3.00 x10 ⁻⁴ ±2.95 x10 ⁻⁴	1.23 x10 ⁻³ ±1.48 x10 ⁻³	1.01 x10 ⁻² ±1.48 x10 ⁻²

Table S11. Mass change of cBOD₅ observed in different components of the treatment system. The +ve values indicate mass losses from the system component.

	cBOD ₅ mass in (mg)	cBOD ₅ mass out (mg)	cBOD ₅ mass change (mg)	cBOD ₅ mass change (Kg)	cBOD ₅ mass % change
Overall treatment system	22388542	361100	22027443	22.03	98.39
Cell 2	22388542	12029514	10359029	10.36	46.27
Cell 3	12029514	3510693	8518820	8.52	70.82
Cell 4	3510693	361100	3149594	3.15	89.71