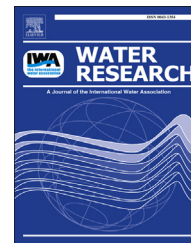


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# Assessing potential modifications to the activated sludge process to improve simultaneous removal of a diverse range of micropollutants

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## ABSTRACT

It is proposed that wastewater treatment facilities meet legislated discharge limits for a range of micropollutants. However, the heterogeneity of these micropollutants in wastewaters make removal difficult to predict since their chemistry is so diverse. In this study, a range of organic and inorganic micropollutants known to be preferentially removed via different mechanisms were selected to challenge the activated sludge process (ASP) and determine its potential to achieve simultaneous micropollutant removal. At a fixed hydraulic retention time (HRT) of 8 h, the influence of an increase in solids retention time (SRT) on removal was evaluated. Maximum achievable micropollutant removal was recorded for all chemicals (estrogens, nonylphenolics and metals) at the highest SRT studied (27 days). Also, optimisation of HRT by extension to 24 h further augmented organic biodegradation. Most notable was the enhancement in removal of the considerably recalcitrant synthetic estrogen 17 $\alpha$ -ethinylestradiol which increased to 65  $\pm$  19%. Regression analysis indicates that this enhanced micropollutant behaviour is ostensibly related to the concomitant reduction in food: microorganism ratio. Interestingly, extended HRT also initiated nonylphenol biodegradation which has not been consistently observed previously in real wastewaters. However, extending HRT increased the solubilisation of particulate bound metals, increasing effluent aqueous metals concentrations (i.e., 0.45  $\mu$ m filtered) by >100%. This is significant as only the aqueous metal phase is to be considered for environmental compliance. Consequently, identification of an optimum process condition for generic micropollutant removal is expected to favour a more integrated approach where upstream process unit optimisation (i.e., primary sedimentation) is demanded to reduce loading of the particle bound metal phase onto the ASP, thereby enabling longer HRT in the ASP to be considered for optimum removal of organic micropollutants.

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## 1. Introduction

Current operations of secondary wastewater treatment works (WwTWs) do not enable compliance to Environmental Quality Standards (EQSs) and proposed legislative targets for a wide variety of micropollutants (Gardner et al., 2012). However, the environmental and economic cost of implementing and operating advanced processes underpins the need to optimise these secondary processes (Jones et al., 2007). Activated sludge is a widely used secondary process with a proven ability to remove a variety of micropollutants (Clara et al., 2005; Koh et al., 2009; Radjenovic et al., 2009; McAdam et al., 2010, 2011; Petrie et al., 2013a). Micropollutant removal by activated sludge can be attributed mostly to sorption and biodegradation mechanisms (Joss et al., 2004; Andersen et al., 2005; Langford et al., 2005; Radjenovic et al., 2009; Petrie et al., 2013a). Therefore measuring process performance and optimising operation is needed on micropollutants which exhibit differing susceptibility to these mechanisms for removal. Natural and synthetic steroid estrogens, nonylphenolic surfactants (NPx) and metals encompass such diversity. These are describable as biodegradable and refractory organics (Joss et al., 2004; Andersen et al., 2005; Clara et al., 2005; Petrie et al., 2013b, submitted for publication) and non-biodegradable inorganics (Santos et al., 2010), respectively. These exhibit a broad range of physico-chemical properties (Table S1) which contributes to their differing fate and behaviour during wastewater treatment. For example, hydrophobicity ( $\log K_{ow}$ ) can be used as a reasonable predictor for sorption behaviour of estrogens (Petrie et al., 2014). Nevertheless, biodegradation is accepted as their major removal pathway (Andersen et al., 2005; Petrie et al., 2014). The removal of NPx is more complex as this family of chemicals possess a variety of physico-chemical properties and breakdown pathways. The relatively hydrophilic long chained NPx's are highly susceptible to biological attack (McAdam et al., 2011). However, their biotransformation can result in the formation of shorter chained intermediates and NP (Petrie et al., 2013b). These chemicals have  $\log K_{ow}$ 's > 5 (Table S1) and a relatively high propensity to sludge partitioning is expected (Byrns, 2001). In contrast, metals rely entirely on partitioning within the activated sludge matrix for removal. This is considered to be driven by three major processes which are; physical entrapment of insoluble metals, binding of soluble metals to bacterial walls and extracellular polymers, and active cellular uptake by bacterial cells (Ziolko et al., 2011).

Micropollutant removal is considered to be influenced by activated sludge process variables, solids retention time (SRT) and hydraulic retention time (HRT) (Svenson et al., 2003; Clara et al., 2005; Johnson et al., 2005; Hamid and Eskicioglu, 2012; Maeng et al., 2013). Despite a large volume of research undertaken on this subject area, it remains unclear whether operation can be optimised to achieve maximum removals of all micropollutants simultaneously. To illustrate, an extended SRT ( $\geq 10$  days) is considered necessary to augment removal of those biodegradable micropollutants (Clara et al., 2005; McAdam et al., 2010). This is thought to enable the enrichment of a more diverse bacterial community more capable of organic micropollutant biodegradation. However, such

conditions are hypothesised to be detrimental to metals removal (Santos et al., 2010). It is anticipated that metals are solubilised by chelators produced by biomasses at SRTs >10 days and are therefore not available for removal within the activated sludge matrix (Santos et al., 2010). Furthermore, there is a paucity of information on the impact of HRT to micropollutant removal. It can be postulated that longer HRTs (and contact time) enable greater biodegradation of the organic micropollutants. Previous research in this field has traditionally focussed on the broad comparison of different full-scale activated sludge plants (ASPs) operating at various SRT and HRTs (Svenson et al., 2003; Clara et al., 2005; Johnson et al., 2005; Koh et al., 2009; McAdam et al., 2010, 2011). However, full-scale processes tend to suffer from poor process control resulting in a dynamic system with considerable variation in both SRT and HRT. To demonstrate, a full-scale ASP designed for operation at 10 day SRT and 8 h HRT was subject to influent flow variations of between 50 and 600 m<sup>3</sup> h<sup>-1</sup> (Aboobakar et al., 2013) which, based on a simple solids mass balance, implies an estimated SRT range of between 4 and 20 days. Consequently, to overcome the limitations of dynamic process conditions observed at full scale, this study utilised a pilot-scale ASP to enable good process control and circumvent variations in receiving sewage composition and flow. This allowed SRT and HRT to be focused on separately and their individual impact to micropollutant removal to be assessed. The pilot-scale ASP was operated at: (i) 3, 10 and 27 day SRTs whilst at a constant HRT (8 h) and (ii) 8, 16 and 24 h HRTs at a constant SRT (27 days). To measure ASP performance for the removal of a wide range of micropollutants of varied chemistry and preferred removal mechanisms under this range of operating conditions - steroid estrogens, NPx's and metals were monitored. To our knowledge this is the first study which has closely controlled ASP operation and measured the impact to the removal of such a diverse range of chemicals simultaneously in real wastewater.

## 2. Materials and methods

### 2.1. Chemicals

Estrogen standards (>98% purity); estrone (E1), 17 $\beta$ -estradiol (E2), estriol (E3), 17 $\alpha$ -ethinylestradiol (EE2) and estrone sulphate (E1-3S) were purchased from Sigma Aldrich (Dorset, UK). Deuterated internal standards; E1-d<sub>4</sub>, E2-d<sub>5</sub>, E3-d<sub>3</sub>, EE2-d<sub>4</sub> and E1-3S-d<sub>4</sub> sulfate were obtained from QMX Laboratories (Thaxted, UK). Technical 4-NP, 4-nonylphenol-monoethoxylate (NP<sub>1</sub>EO), the diethoxylate compound (NP<sub>2</sub>EO) and the longer chain NPEOs (NP<sub>3-12</sub>EO) were purchased from Sigma Aldrich (Dorset, UK). Long chain NPEOs were purchased as the technical mixtures CO210, CO520 and CO720. Nonylphenoxy acetic acid (NP<sub>1</sub>EC) was obtained from QMX laboratories (Thaxted, UK). Single element metal solutions of zinc (Zn), copper (Cu), lead (Pb), nickel (Ni), cadmium (Cd) and rhodium (internal standard), and OPTIMA trace metal grade nitric acid were obtained from Fisher Scientific (Leicestershire, UK). The high performance liquid chromatography grade solvents; acetone, methanol, dichloromethane, ethylacetate and hexane were purchased from Rathburn Chemicals

(Walkerburn, UK). Ammonium hydroxide was ACS grade and obtained from Sigma Aldrich (Dorset, UK) and ultra-pure water of 18.2 M $\Omega$  quality (Elga, Marlow, UK) was used in the preparation of mobile phases. Chemical oxygen demand (COD), ammoniacal nitrogen, nitrate, nitrite and total nitrogen proprietary cell test kits were purchased from VWR International (Leicestershire, UK).

## 2.2. Pilot plant operation

A pilot-scale ASP was sited at a WwTWs in the east of England (3000 population equivalent) and consisted of a 0.18 m<sup>3</sup> primary sedimentation tank, a 0.36 m<sup>3</sup> aerated basin and a 0.10 m<sup>3</sup> final clarifier (Petrie et al., 2014) (Fig. 1). The plant was seeded with biomass from a full-scale ASP (280,000 population equivalent) and operated with municipal crude sewage containing indigenous concentrations of all micropollutants (Petrie et al., 2013b, 2013c). Less than 5% of the crude sewage was attributed to industrial inputs from a local airfield. The influent flow rate of the pilot plant was controlled to achieve a constant HRT. Return activated sludge (RAS) was 0.55 of the influent flow in all studies. Solids retention time was controlled by daily wastage of sludge (waste activated sludge, WAS) from the base of the final clarifier following correction for loss of effluent suspended solids. The system was operated for at least three SRTs prior to monitoring at each different condition to ensure continuity of SRT and HRT performance was established. Once under steady state conditions, sampling was undertaken over seven consecutive days (Monday to Sunday) at varying times between 7:00 and 19:00 h. Samples (~10 L – to obtain suitable quantities of suspended solids for particulate analysis) were collected in 2.5 L borosilicate glass vessels with Teflon lined caps for the determination of organic micropollutants and sanitary determinands. Samples for metals analysis were collected using acid washed 50 ml plastic centrifuge tubes. Grab sampling was employed due to the well-known problems of chemical stability whilst using composite samplers (Baker and Kasprzyk-Hordern, 2013). Upon collection, samples were transported to the laboratory (within 10 min) and processed immediately. During the seven day sampling campaigns which were performed over a two year period, no significant rainfall was experienced. The impact of SRT was investigated by maintaining a constant 8 h HRT over the secondary treatment stage. Solid retention times of 3, 10 and 27 days were assessed. In HRT studies the SRT was maintained at 27 days and HRTs of 8, 16 and 24 h examined.

## 2.3. Sanitary determine and analysis

Chemical oxygen demand, soluble COD, ammoniacal nitrogen, nitrate, nitrite and total nitrogen were determined using proprietary cell test kits (VWR International, Leicestershire, UK) and subsequent detection by spectrophotometry. The aqueous phase of wastewater was obtained by filtration through a 1.2  $\mu$ m filter (Whatman, Maidstone, UK). Suspended solids, volatile suspended solids (VSS) and biochemical oxygen demand (BOD) were determined using standard methods (APHA, 1998). Biomass of each operational condition was characterised by separating extracellular polymeric substance (EPS) and soluble micro products fractions (Le-Clech et al.,

2006). Briefly, 50 ml of biomass was centrifuged at 1500 $\times$  *g* for 5 min and the supernatant filtered (0.2  $\mu$ m) to obtain the SMP fraction. The solid fraction was re-suspended in 50 ml of deionised water and placed in an oven at 105 °C for 1 h. The extracted EPS was obtained by allowing the sample to cool to room temperature, centrifuging at 1500 $\times$  *g* for 5 min and filtering (0.2  $\mu$ m). Protein and carbohydrate concentrations were quantified using the phenol–sulphuric acid method (Zhang et al., 1999) and modified Lowry method (Frølund et al., 1995) respectively. Charge (as zeta potential) was determined using a Zetasizer 2000 (Malvern Instruments LTD, Worcestershire, UK) whereas activated sludge floc size was measured using a Mastersizer 2000 (Malvern Instruments LTD, Worcestershire, UK).

## 2.4. Micropollutant analysis

### 2.4.1. Steroid estrogens

For all samples collected (settled sewage, RAS/WAS and final effluent), aqueous and particulate phases were separated by centrifugation and filtration (Petrie et al., 2013c). The separated aqueous and particulate phases were then analysed separately using a three stage extraction and clean up procedure. Quantification was by ultra-performance liquid chromatography tandem mass spectrometry (UPLC-MS/MS) as previously described (Petrie et al., 2013c). A triple quadrupole was utilised and two multiple reaction monitoring (MRM) transitions were monitored per compound. Deuterated surrogates were used to compensate for any loss of analytes during sample preparation. The method achieved aqueous and particulate method detection limits (MDLs) of  $5.0 \times 10^{-5}$  to  $1.7 \times 10^{-4}$   $\mu$ g l<sup>-1</sup> and  $2.5 \times 10^{-3}$  to  $8.9 \times 10^{-3}$   $\mu$ g g<sup>-1</sup>, respectively. Steroid estrogen recoveries ranged from 46.9 to 114.3% for the aqueous and particulate phases.

### 2.4.2. Nonylphenolics

Similarly, all samples for nonylphenolic analysis were separated into aqueous and particulate phases and analysed individually. Analysis utilised a single stage solid phase extraction and quantitation by UPLC-MS/MS which has been detailed elsewhere (Petrie et al., 2013b). The triple quadrupole monitored two MRMs (quantifier and qualifier) for each chemical. Nonylphenolic MDLs ranged from  $1.4 \times 10^{-3}$  to  $4.5 \times 10^{-2}$   $\mu$ g l<sup>-1</sup> for aqueous phases and from  $1.4 \times 10^{-3}$  to  $3.9 \times 10^{-2}$   $\mu$ g g<sup>-1</sup> for particulate phases. Recoveries for all nonylphenolic chemicals from the aqueous and particulate phases of wastewaters ranged from 40.3 to 102.1% during the studies.

### 2.4.3. Metals

Sample preparation for metals analysis was similar to that reported by Santos et al. (2010). Briefly, samples were filtered (0.45  $\mu$ m) to obtain the aqueous phase. Analysis of the unfiltered sample was also performed to determine total concentration. Particulate concentration was then calculated from the difference of the total and aqueous concentrations. Metals analysis was also undertaken on receiving crude sewage to monitor removal over primary sedimentation due to their relatively high affinity to suspended particulate matter. Samples were analysed by an ELAN 9000 inductively coupled

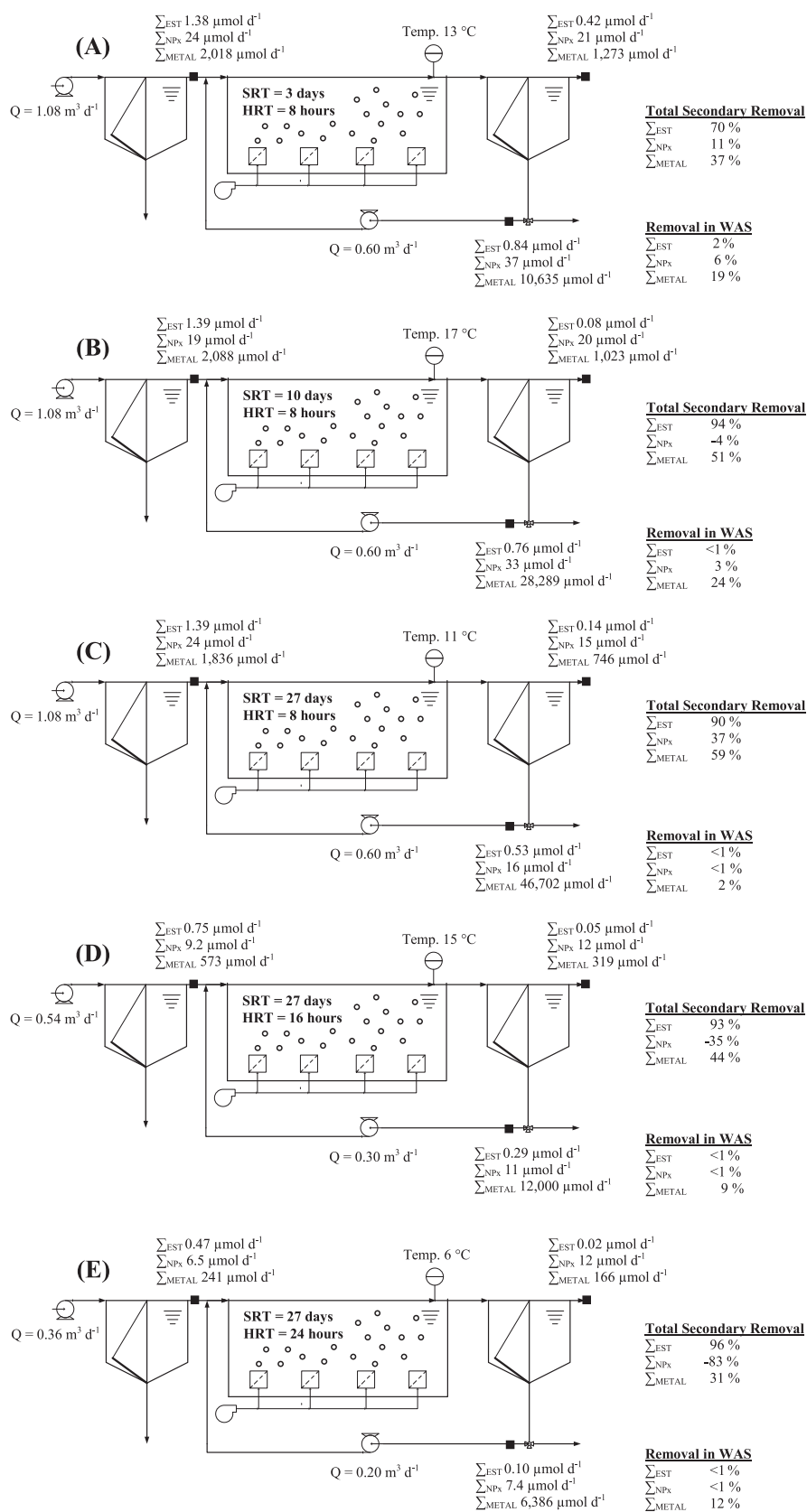


Fig. 1 – Mass balance data for steroid estrogens, NPx and metals at varying operational conditions (A, 3 day SRT 8 h HRT; B, 10 day SRT 8 h HRT; C, 27 day SRT 8 h HRT; D, 27 day SRT 16 h HRT; E, 27 day SRT 24 h HRT; ■, sampling points).

Table 1 – Micropollutant quantitative information reported as mean for SRT and HRT pilot plant studies (n = 7).

Chemical	Crude sewage ( $\mu\text{g l}^{-1}$ )			SRT (d)	HRT (h)	Settled sewage ( $\mu\text{g l}^{-1}$ )			Primary removal <sup>a</sup> (%)	RAS ( $\mu\text{g l}^{-1}$ )			Final effluent ( $\mu\text{g l}^{-1}$ )			Secondary removal <sup>b</sup> (%)		
	Aq.	Part.	Total			Aq.	Part.	Total		Aq.	Part.	Total	Aq.	Part.	Total	Aq.	Part.	Total
Zn	11.2	80.1	91.3	3	8	11.6	61.5	73.1	14 <sup>c</sup>	15.4	553	568	16.5	29.8	46.3	-42	52	38
						25.1	63.4	88.5		69.5	$1.50 \times 10^3$	$1.57 \times 10^3$	35.2	8.90	44.1	-40	86	50
						16.4	58.7	75.1		39.9	$2.82 \times 10^3$	$2.86 \times 10^3$	31.4	6.60	38.0	-91	89	49
						8.27	32.9	41.2	55	25.6	$1.04 \times 10^3$	$1.07 \times 10^3$	22.6	5.10	27.7	-173	85	33
						9.03	16.8	25.8	72	20.4	921	941	19.2	2.90	22.1	-113	83	14
Cu	1.50	44.3	45.8	3	8	2.90	42.7	45.6	16 <sup>c</sup>	2.37	546	548	3.90	24.4	28.3	-34	43	38
						2.69	33.5	36.2		5.50	$1.39 \times 10^3$	$1.39 \times 10^3$	4.40	12.5	16.9	-64	63	53
						1.72	32.3	34.0		3.82	$2.13 \times 10^3$	$2.13 \times 10^3$	2.40	3.80	6.20	-40	88	82
						1.20	25.8	27.0	41	4.41	$1.42 \times 10^3$	$1.42 \times 10^3$	4.23	6.07	10.3	-253	76	62
						1.45	15.7	17.1	63	4.11	$1.10 \times 10^3$	$1.10 \times 10^3$	4.21	3.28	7.48	-190	79	56
Pb	0.673	2.97	3.64	3	8	0.750	1.39	2.14	70 <sup>c</sup>	0.500	24.0	24.5	0.600	0.880	1.48	15	51	31
						0.290	0.720	1.01		0.500	78.9	79.4	0.400	0.317	0.717	-110	56	29
						<0.116	0.111	0.227		0.150	60.4	60.6	<0.116	-	<0.116	-	-	>49
						0.260	0.553	0.813	78	0.470	46.8	47.3	0.430	-	0.426	-65	23	48
						0.550	0.220	0.770	78	0.620	10.2	10.8	0.500	$5.60 \times 10^{-2}$	0.556	9	75	28
Cd	-	-	-	3	8	$<9.56 \times 10^{-2}$	0.107	0.203	-	$<9.56 \times 10^{-2}$	1.84	1.94	0.110	$6.00 \times 10^{-3}$	0.116	-15	99	43
						$<9.56 \times 10^{-2}$	$6.54 \times 10^{-2}$	0.161		$<9.56 \times 10^{-2}$	6.15	6.25	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	-	>41
						$<9.56 \times 10^{-2}$	$8.74 \times 10^{-2}$	0.183		$<9.56 \times 10^{-2}$	3.74	3.84	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	-	>48
						$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	2.60	2.70	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	-	-
						$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	1.08	1.18	$<9.56 \times 10^{-2}$	-	$<9.56 \times 10^{-2}$	-	-	-
Ni	-	-	-	3	8	0.580	0.740	1.32	-	0.530	15.6	16.1	0.400	0.591	0.991	31	20	25
						0.130	0.207	0.207		0.300	52.2	52.5	0.140	$3.60 \times 10^{-2}$	0.176	-8	83	15
						0.490	0.347	0.837		1.87	44.7	46.6	0.490	0.167	0.657	0	52	22
						<0.120	-	<0.120	-	<0.120	11.4	11.5	<0.120	-	<0.120	-	-	-
						<0.120	-	<0.120	-	<0.120	4.20	4.32	<0.120	-	<0.120	-	-	-
E1	-	-	-	3 <sup>d</sup>	8	$8.91 \times 10^{-2}$	$2.60 \times 10^{-3}$	$9.17 \times 10^{-2}$	-	0.104	$8.60 \times 10^{-2}$	0.190	$7.40 \times 10^{-2}$	$5.02 \times 10^{-4}$	$7.45 \times 10^{-2}$	17	81	19
						$9.56 \times 10^{-2}$	$5.40 \times 10^{-3}$	0.101		$9.01 \times 10^{-2}$	0.104	0.194	$6.72 \times 10^{-3}$	$4.20 \times 10^{-4}$	$7.14 \times 10^{-3}$	93	92	93
						$9.12 \times 10^{-2}$	$2.80 \times 10^{-3}$	$9.40 \times 10^{-2}$		$6.51 \times 10^{-2}$	$6.50 \times 10^{-2}$	0.131	$1.46 \times 10^{-2}$	$1.98 \times 10^{-4}$	$1.48 \times 10^{-2}$	84	93	84
						0.100	$1.98 \times 10^{-3}$	0.102		$8.60 \times 10^{-2}$	$8.20 \times 10^{-2}$	0.168	$1.42 \times 10^{-2}$	$3.98 \times 10^{-4}$	$1.46 \times 10^{-2}$	86	80	86
						$7.17 \times 10^{-2}$	$2.01 \times 10^{-3}$	$7.37 \times 10^{-2}$		$3.56 \times 10^{-3}$	$5.84 \times 10^{-2}$	$6.20 \times 10^{-2}$	$4.90 \times 10^{-3}$	$2.10 \times 10^{-5}$	$4.92 \times 10^{-3}$	93	99	93
E2	-	-	-	3 <sup>d</sup>	8	$2.58 \times 10^{-2}$	$4.30 \times 10^{-3}$	$3.01 \times 10^{-2}$	-	$3.38 \times 10^{-2}$	$7.22 \times 10^{-2}$	0.106	$8.70 \times 10^{-3}$	$1.01 \times 10^{-3}$	$9.71 \times 10^{-3}$	66	77	68
						$2.23 \times 10^{-2}$	$6.20 \times 10^{-3}$	$2.85 \times 10^{-2}$		$1.30 \times 10^{-2}$	$7.62 \times 10^{-2}$	$8.92 \times 10^{-2}$	$2.21 \times 10^{-3}$	$7.11 \times 10^{-4}$	$2.92 \times 10^{-3}$	90	89	90
						$2.67 \times 10^{-2}$	$3.70 \times 10^{-3}$	$3.04 \times 10^{-2}$		$2.61 \times 10^{-3}$	$5.57 \times 10^{-2}$	$5.83 \times 10^{-2}$	$2.22 \times 10^{-3}$	$1.41 \times 10^{-4}$	$2.36 \times 10^{-3}$	92	96	92
						$3.32 \times 10^{-2}$	$7.49 \times 10^{-3}$	$4.07 \times 10^{-2}$		$1.11 \times 10^{-2}$	$2.96 \times 10^{-2}$	$4.07 \times 10^{-2}$	$3.18 \times 10^{-3}$	$1.48 \times 10^{-4}$	$3.33 \times 10^{-3}$	90	98	92
						$3.04 \times 10^{-2}$	$1.13 \times 10^{-2}$	$4.17 \times 10^{-2}$		$2.10 \times 10^{-3}$	$3.41 \times 10^{-2}$	$3.62 \times 10^{-2}$	$2.12 \times 10^{-3}$	$4.99 \times 10^{-5}$	$2.17 \times 10^{-3}$	93	99	95
E3	-	-	-	3 <sup>d</sup>	8	0.219	$1.12 \times 10^{-3}$	0.220	-	$1.49 \times 10^{-2}$	$2.81 \times 10^{-2}$	$4.30 \times 10^{-2}$	$1.66 \times 10^{-2}$	$3.96 \times 10^{-4}$	$1.70 \times 10^{-2}$	92	65	92
						0.212	$2.04 \times 10^{-3}$	0.214		$8.60 \times 10^{-4}$	$2.11 \times 10^{-2}$	$2.20 \times 10^{-2}$	$6.14 \times 10^{-3}$	$6.09 \times 10^{-4}$	$6.75 \times 10^{-3}$	97	70	97
						0.203	$1.98 \times 10^{-3}$	0.205		$1.65 \times 10^{-2}$	$8.40 \times 10^{-3}$	$2.29 \times 10^{-2}$	$5.83 \times 10^{-3}$	$3.92 \times 10^{-4}$	$6.22 \times 10^{-3}$	97	80	97
						0.228	$6.11 \times 10^{-3}$	0.234		$9.39 \times 10^{-4}$	$1.66 \times 10^{-2}$	$1.75 \times 10^{-2}$	$3.83 \times 10^{-3}$	$8.23 \times 10^{-5}$	$3.91 \times 10^{-3}$	98	99	98
						0.232	$1.02 \times 10^{-3}$	0.233		$2.70 \times 10^{-3}$	$1.93 \times 10^{-2}$	$2.20 \times 10^{-2}$	$4.08 \times 10^{-3}$	$5.11 \times 10^{-5}$	$4.13 \times 10^{-3}$	98	95	98



EE2	–	–	–	3 <sup>d</sup>	8	$6.10 \times 10^{-4}$	$1.17 \times 10^{-3}$	$1.78 \times 10^{-3}$	–	$3.00 \times 10^{-4}$	$3.26 \times 10^{-2}$	$3.29 \times 10^{-2}$	$8.65 \times 10^{-4}$	$3.95 \times 10^{-4}$	$1.26 \times 10^{-3}$	–40	66	30
				10 <sup>d</sup>	8	$5.10 \times 10^{-4}$	$3.96 \times 10^{-4}$	$9.06 \times 10^{-4}$	–	$4.30 \times 10^{-4}$	$1.99 \times 10^{-2}$	$2.03 \times 10^{-2}$	$6.30 \times 10^{-4}$	$1.70 \times 10^{-5}$	$6.47 \times 10^{-4}$	–24	96	29
				27 <sup>d</sup>	8	$1.04 \times 10^{-3}$	$4.51 \times 10^{-4}$	$1.49 \times 10^{-3}$	–	$3.40 \times 10^{-4}$	$1.45 \times 10^{-2}$	$1.48 \times 10^{-2}$	$7.00 \times 10^{-4}$	$1.80 \times 10^{-4}$	$8.80 \times 10^{-4}$	33	60	41
				27	16	$3.99 \times 10^{-4}$	$1.73 \times 10^{-4}$	$5.72 \times 10^{-4}$	–	$6.00 \times 10^{-5}$	$1.12 \times 10^{-2}$	$1.13 \times 10^{-2}$	$1.64 \times 10^{-4}$	$9.20 \times 10^{-5}$	$2.56 \times 10^{-4}$	59	47	55
				27	24	$2.68 \times 10^{-4}$	$1.30 \times 10^{-4}$	$3.98 \times 10^{-4}$	–	$8.00 \times 10^{-5}$	$1.32 \times 10^{-2}$	$1.33 \times 10^{-2}$	$1.22 \times 10^{-4}$	$1.80 \times 10^{-5}$	$1.40 \times 10^{-4}$	55	86	65
E <sub>1–3</sub> S	–	–	–	3	8	$2.21 \times 10^{-2}$	$3.01 \times 10^{-4}$	$2.24 \times 10^{-2}$	–	$7.60 \times 10^{-3}$	$1.07 \times 10^{-2}$	$1.83 \times 10^{-2}$	$4.63 \times 10^{-3}$	$9.06 \times 10^{-5}$	$4.72 \times 10^{-3}$	79	70	79
				10	8	$2.36 \times 10^{-2}$	$1.95 \times 10^{-4}$	$2.38 \times 10^{-2}$	–	$8.07 \times 10^{-3}$	$2.05 \times 10^{-2}$	$2.86 \times 10^{-2}$	$3.42 \times 10^{-3}$	$2.96 \times 10^{-5}$	$3.45 \times 10^{-3}$	86	85	85
				27	8	$4.08 \times 10^{-2}$	$1.99 \times 10^{-4}$	$4.10 \times 10^{-2}$	–	$1.65 \times 10^{-2}$	$6.50 \times 10^{-3}$	$2.33 \times 10^{-2}$	$1.45 \times 10^{-2}$	$1.96 \times 10^{-4}$	$1.47 \times 10^{-2}$	64	2	64
				27	16	$1.96 \times 10^{-2}$	$1.02 \times 10^{-4}$	$1.97 \times 10^{-2}$	–	$2.23 \times 10^{-2}$	$5.31 \times 10^{-3}$	$2.76 \times 10^{-2}$	$5.61 \times 10^{-3}$	$2.11 \times 10^{-5}$	$5.63 \times 10^{-3}$	71	79	71
				27	24	$2.40 \times 10^{-2}$	$2.04 \times 10^{-4}$	$2.42 \times 10^{-2}$	–	$4.90 \times 10^{-3}$	$6.21 \times 10^{-3}$	$1.11 \times 10^{-2}$	$4.36 \times 10^{-3}$	$2.11 \times 10^{-5}$	$4.38 \times 10^{-3}$	82	90	82
NP	–	–	–	3	8	1.02	0.560	1.58	–	0.653	1.84	2.50	1.29	0.444	1.74	–26	21	–10
				10	8	0.479	0.583	1.06	–	0.641	2.68	3.32	0.876	0.503	1.38	–83	14	–30
				27	8	1.13	0.534	1.66	–	0.669	0.920	1.59	1.40	0.360	1.76	–23	33	–6
				27	16	0.225	$2.26 \times 10^{-2}$	0.248	–	$<9.00 \times 10^{-3}$	1.16	1.17	$<9.00 \times 10^{-3}$	$6.42 \times 10^{-2}$	$7.32 \times 10^{-2}$	>96	–184	70
				27	24	0.533	$9.56 \times 10^{-2}$	0.629	–	0.247	0.794	1.04	0.231	$3.37 \times 10^{-3}$	0.234	57	96	63
NP <sub>1–3</sub> EC	–	–	–	3	8	0.144	$1.11 \times 10^{-2}$	0.155	–	1.44	7.09	8.54	1.30	$2.30 \times 10^{-2}$	1.32	–803	–107	–751
				10	8	0.144	$2.20 \times 10^{-2}$	0.166	–	1.42	4.98	6.41	2.43	$1.10 \times 10^{-2}$	2.44	–1588	49	–1370
				27	8	0.168	$1.01 \times 10^{-2}$	0.178	–	0.362	$3.04 \times 10^{-3}$	0.365	0.554	$1.16 \times 10^{-3}$	0.555	–230	81	–212
				27	16	3.11	$1.04 \times 10^{-2}$	3.12	–	8.55	0.389	8.94	6.92	$1.43 \times 10^{-3}$	6.92	–123	86	–122
				27	24	2.52	$6.00 \times 10^{-3}$	2.53	–	8.71	0.445	9.16	10.1	$2.10 \times 10^{-3}$	10.1	–301	65	–297
NP <sub>1–3</sub> EO	–	–	–	3	8	1.60	0.929	2.53	–	1.70	3.78	5.49	1.67	0.124	1.80	–4	87	30
				10	8	1.25	0.909	2.16	–	0.634	4.39	5.02	0.915	0.224	1.14	27	75	48
				27	8	1.22	0.879	2.10	–	0.463	4.48	4.94	0.729	0.303	1.03	40	66	52
				27	16	0.296	$2.11 \times 10^{-3}$	0.298	–	$2.20 \times 10^{-2}$	$1.09 \times 10^{-4}$	$2.20 \times 10^{-2}$	$1.94 \times 10^{-2}$	$6.99 \times 10^{-4}$	$2.01 \times 10^{-2}$	93	67	93
				27	24	0.395	0.153	0.548	–	0.132	0.280	0.411	$1.98 \times 10^{-2}$	$5.10 \times 10^{-4}$	$2.03 \times 10^{-2}$	95	99	96
NP <sub>4–12</sub> EO	–	–	–	3	8	2.24	0.504	2.74	–	0.165	2.17	2.33	0.408	$8.62 \times 10^{-2}$	0.494	82	83	82
				10	8	1.95	0.736	2.68	–	0.157	1.61	1.77	0.221	$4.79 \times 10^{-2}$	0.269	89	93	89
				27	8	3.18	0.369	3.55	–	0.103	0.356	0.460	0.195	$7.28 \times 10^{-2}$	0.268	94	79	92
				27	16	2.65	0.295	2.95	–	$3.92 \times 10^{-2}$	1.01	1.05	0.105	0.123	0.229	96	58	93
				27	24	2.56	0.510	3.07	–	0.156	0.932	1.09	0.223	$2.61 \times 10^{-2}$	0.245	92	95	92

Key: Aq., aqueous; Part., particulate; SRT, solids retention time; HRT, hydraulic retention time; RAS, return activated sludge; –, not determined.

<sup>a</sup> Primary removal (%) = crude sewage – settled sewage/crude sewage 100%.

<sup>b</sup> Secondary removal (%) = settled sewage – final effluent/settled sewage 100%.

<sup>c</sup> Mean primary removal from three sampling campaigns.

<sup>d</sup> Petrie et al., 2014.

plasma-mass spectrometer (Perkin Elmer, Beaconsfield, UK). Metal MDLs ranged from  $6.6 \times 10^{-2}$  to  $0.5 \mu\text{g l}^{-1}$ . Total and aqueous metal recoveries ranged from 73.1 to 106.9%. Full descriptions of all micropollutant methods are available in Supplementary Material.

### 3. Results and discussion

#### 3.1. Impact of ASP operation on micropollutant removal

##### 3.1.1. Steroid estrogens

The SRT range studied broadly encompassed typical full-scale operations (Joss et al., 2004; Clara et al., 2005; Johnson et al., 2005; Koh et al., 2009; McAdam et al., 2010, 2011). Process performance at each operating condition studied was ascertained by calculating micropollutant removal according to:

$$\text{Secondary removal(\%)} = \frac{\text{SS} - \text{FE}}{\text{SS}} \cdot 100\% \quad (1)$$

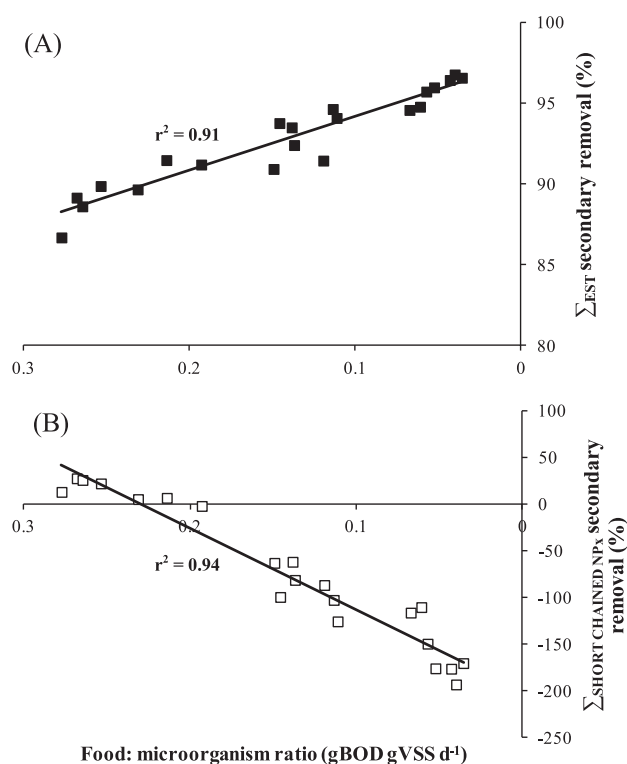
Here SS is the concentration of a given micropollutant in settled sewage and FE is the concentration in final effluent. Removal of total estrogens ( $\sum_{\text{EST}}$ , i.e., sum of E1, E2, E3 and EE2 accounting for aqueous and particulate concentrations) at the 3 day SRT condition (8 h HRT) was  $70 \pm 7\%$  (Fig. 1). Increased SRT (i.e., 10 and 27 days) achieved augmented removals. However, no improvement in  $\sum_{\text{EST}}$  removal was observed when SRT was increased from 10 to 27 days. Removal of  $\sum_{\text{EST}}$  was  $94 \pm 1\%$  and  $90 \pm 2\%$  respectively. Higher  $\sum_{\text{EST}}$  removals here were driven by improved natural estrogen (E1, E2 and E3) removals (Table 1). Removals of E1, E2 and E3 individually were  $\geq 84\%$  for 10 and 27 day SRTs. These are within the range of removals previously reported for various full-scale nitrifying processes (Joss et al., 2004; Clara et al., 2005; Johnson et al., 2005; Koh et al., 2009; McAdam et al., 2010). On the other hand, no improvement in EE2 removal was observed at the 10 or 27 day SRT. Mean removals were in the range 29–41% (Table 1). A broad range of EE2 removals by activated sludge treatment have been observed in the literature, ranging from 38 to 98% (Joss et al., 2004; Clara et al., 2005; Koh et al., 2009; McAdam et al., 2010), with no clear relationship apparent between process operation and removal. Despite increasing SRT, the quantity of  $\sum_{\text{EST}}$  within RAS reduced (Fig. 1). This, coupled with low estrogen removal in WAS (Fig. 1) infers the majority of removal during activated sludge treatment is attributed to biodegradation (Andersen et al., 2005; Petrie et al., 2014).

Steady state HRTs of 8, 16 and 24 h were monitored to represent typical full-scale (dry weather flow) operational conditions (Johnson et al., 2005; Koh et al., 2009; McAdam et al., 2010, 2011). A fixed SRT of 27 days was employed as this exhibited best overall removal performance previously. Removals of  $\sum_{\text{EST}}$ 's were  $90 \pm 2\%$ ,  $93 \pm 1\%$  and  $96 \pm 2\%$  for 8, 16 and 24 h HRTs, respectively (Fig. 1). Individually, each estrogen (including EE2) showed improving removal with longer HRT. At 24 h HRT, removals of the natural estrogens E1, E2 and E3 were all  $\geq 93\%$  (Table 1). Most notably EE2 exhibited removals of  $65 \pm 19\%$  at the 24 h HRT condition (Table 1). Lengthening HRT of the secondary aerobic treatment stage saw a concomitant decrease in food to microorganism (F: M) ratio. Ratios were  $0.25 \pm 0.03$ ,  $0.13 \pm 0.02$  and  $0.05 \pm 0.01$  gBOD

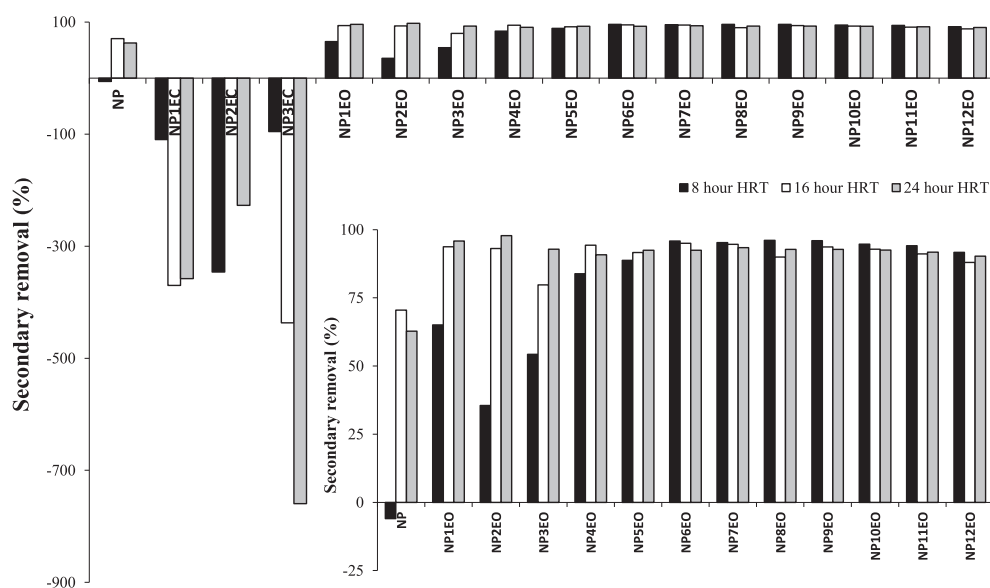
gVSS  $\text{d}^{-1}$  for 8, 16 and 24 h HRTs, respectively. Interestingly, reducing F:M ratio correlated well ( $r^2$  0.91) with increased secondary removal of  $\sum_{\text{EST}}$  (Fig. 2). A lower F: M ratio is suggestive of a substrate limitation which may lead to the biodegradation of less-favoured carbon substrates (e.g., steroid estrogens). Coupled with increased contact time for biodegradation, this may explain the observed improvement in biodegradation at longer HRTs (Fig. 1).

##### 3.1.2. Nonylphenolics

Activated sludge treatment initiated NPEO chain shortening with removals of  $\text{NP}_{4-12}\text{EO} \geq 79\%$  for all pilot plant studies (Table 1). However, removal of total nonylphenolic chemicals ( $\sum_{\text{NPx}}$ , sum of NP,  $\text{NP}_{1-12}\text{EO}$  and  $\text{NP}_{1-3}\text{EC}$  accounting for aqueous and particulate concentrations) during studies investigating SRT was relatively low. At 3 and 10 day SRTs removal of  $\sum_{\text{NPx}}$  was negligible; 6% and  $-4\%$ , respectively (Fig. 1). Reported removals of total NPx load in the literature are typically higher (Koh et al., 2009; McAdam et al., 2011) and removals up to 95% have previously been observed for a full-scale ASP operated at an SRT of 13 days (Koh et al., 2009). This is because these studies report load as mass (i.e.,  $\text{ng l}^{-1}$ ) instead of moles which can overestimate removal for this chemical type (Petrie et al., 2013b). At 27 days SRT removal of  $37 \pm 5\%$  was achieved (Fig. 1). This suggests an improvement across the NPx removal pathway by assimilation and oxidation of breakdown intermediates. Higher  $\sum_{\text{NPx}}$  removal was driven by an improved removal of  $\text{NP}_{1-3}\text{ECs}$  ( $-212 \pm 98\%$ ) (Table 1). The specific difficulty associated with this family of



**Fig. 2 – Significance of the food: microorganism ratio to secondary removal of  $\sum_{\text{EST}}$ 's (A, ■) and short chained NPx (NP +  $\text{NP}_{1-3}\text{EO}$  +  $\text{NP}_{1-3}\text{EC}$ ) (B, □) in HRT studies.**



**Fig. 3 – Secondary removal of individual nonylphenolic chemicals at varying HRT. Inset, individual removals of NP and NP<sub>1–12</sub>EO.**

chemicals is the complexity of biotransformation reactions that this family of chemicals exhibit prior to NP biodegradation. To demonstrate, a number of structural changes (e.g., ethoxylate shortening, oxidations etc) are undertaken before cleavage of the NP ring takes place (Warhurst, 1994; Petrie et al., 2013b). The proposed biotransformation pathway of NPEO's is available in Supplementary Material (Figure S1). In this study, removals of the daughter chemical NP ranged from  $-6\%$  to  $-30\%$  over the three SRTs studied (Table 1). As an independent chemical under controlled conditions, NP is known to be susceptible to biodegradation (Tanghe et al., 1998; Stasinakis et al., 2008), even at low SRT (Stasinakis et al., 2010). This suggests its production during activated sludge treatment by precursor biotransformation here was sufficient to compensate for its removal (i.e., net removal is zero).

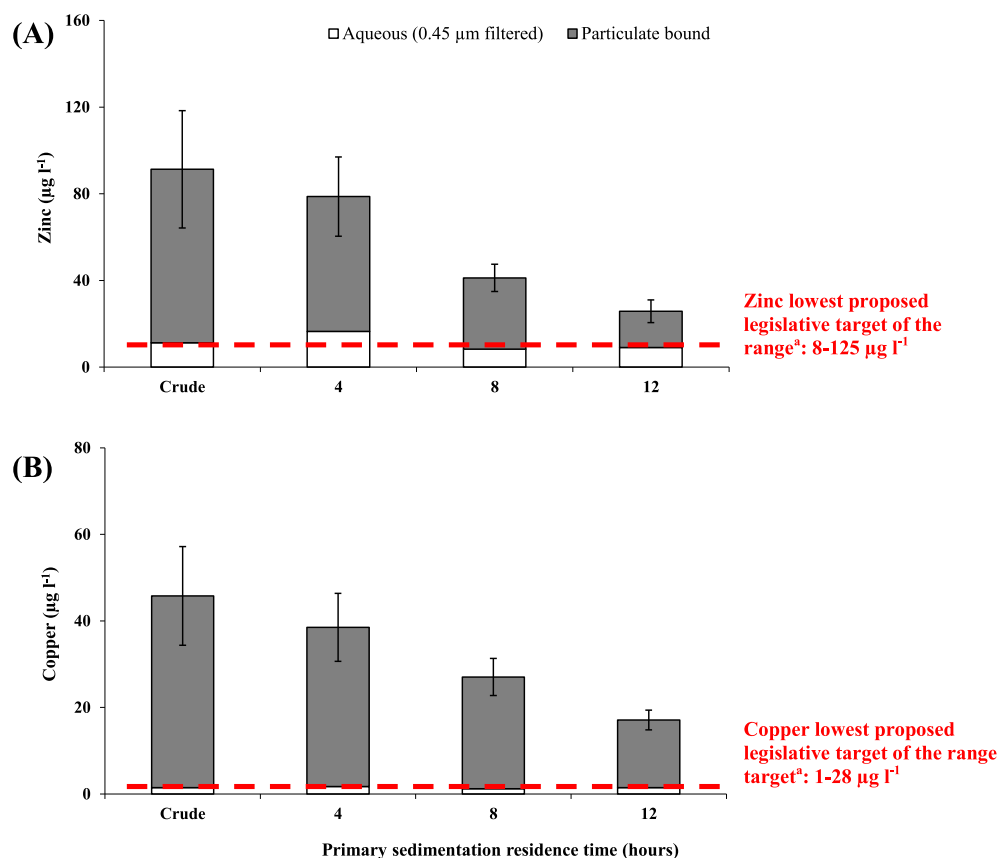
Lengthening HRT resulted in an apparent increase in  $\sum_{\text{NPx}}$  molar concentration with removals of  $37 \pm 5\%$ ,  $-35 \pm 18\%$  and  $-83 \pm 12\%$  observed for 8, 16 and 24 h, respectively (Fig. 1). This illustrates that all chemicals within the NPx chemical family are not encompassed by current analytical methods. For example, longer chained NPEOs (Petrie et al., 2013b) and NPECs (Komori et al., 2006), and carboxylated carboxylates (i.e., NP molecule containing two carboxylic acid functional groups) (Jonkers et al., 2001) can be present. Without analytical reference standards available for their quantitation and inclusion in the nonylphenolic mass balance, knowledge of their fate during treatment and resultant impact to NP removal remains limited. A negative correlation ( $r^2$  0.94) was observed between reduced F:M ratio and short chained NPx removal (i.e., NP + NP<sub>1–3</sub>EO + NP<sub>1–3</sub>EC) (Fig. 2). This was mainly attributed to increased concentrations of NP<sub>1–3</sub>EC observed in final effluents (Table 1). Nevertheless, this counter-intuitively resulted in the greatest removals of NP. Positive removals of  $70 \pm 16\%$  and  $63 \pm 15\%$  were observed at 16 and 24 h HRTs respectively (Fig. 3). Removals of  $>50\%$  have previously been reported in the literature for NP (Nakada et al.,

2006; Qiang et al., 2013), but these have not been supported with parent compound analysis which is needed to better understand pathways of biotransformation. The high removal of all NPEOs observed at 16 and 24 h HRTs (Table 1) indicates that ethoxylate biotransformation was the dominant pathway for the production of NP during ASP treatment. Therefore longer HRT was advantageous as this initiated improved biotransformation across the NPx removal pathway resulting in notable NP removal. It should be noted that the vapour pressure ( $4.39 \times 10^{-5}$  kPa) and Henry's law constant ( $4.30 \times 10^{-6}$  atm m<sup>3</sup>mol<sup>-1</sup>) (Table S1) of NP indicates a semi-volatile nature (Soares et al., 2008). Therefore increased removal by volatilisation at longer HRTs cannot be discounted.

### 3.1.3. Metals

In contrast to the organic micropollutants, total metal ( $\sum_{\text{METAL}}$ , sum of Zn, Cu, Pb, Cd and Ni accounting for aqueous and particulate concentrations) removal increased moderately with each SRT increment. Removals were  $34 \pm 19\%$ ,  $51 \pm 10\%$  and  $59 \pm 11\%$  for 3, 10 and 27 day SRTs respectively (Fig. 1). This was mainly attributed to Cu and Zn as they constituted the majority of metals concentration found in receiving sewage (Table 1). Specifically, Cu removals were improved by increasing SRT and were  $38 \pm 15\%$ ,  $53 \pm 9\%$  and  $82 \pm 4\%$ . In contrast, Ni removals were low and in the range; 15–25%. The difficulty of removing Ni during biological wastewater treatment is well established (Ziolko et al., 2011). Partitioning within the biomass matrix was confirmed as the main removal pathway for metals as increased concentrations were observed in the particulate phase of RAS at each SRT increase (Fig. 1, Table 1). The moderate improvement (8%) in  $\sum_{\text{METAL}}$  removal between the 10 and 27 SRTs coincided with similar MLVSS concentrations (Fig. 1, Table S2). This suggests enhanced metal partitioning within the biomass matrix, likely to be induced by a physiological change to the biomass





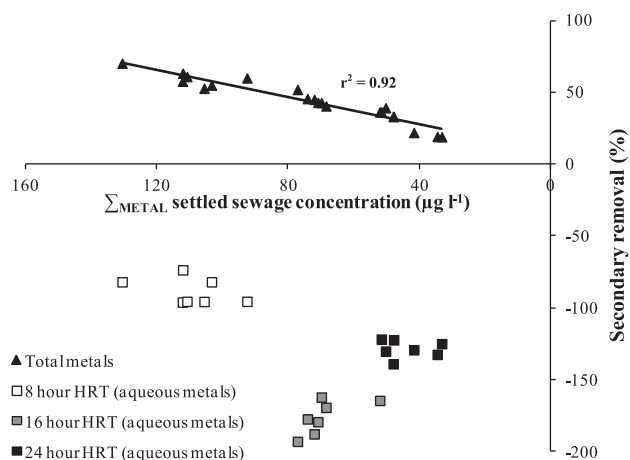
**Fig. 4 – Effect of primary sedimentation tank residence time on Zn (A) and Cu (B) concentration in settled sewage. <sup>a</sup>EQS is applied to the aqueous concentration (i.e., pre-filtered) and is dependent on water hardness (European Commission, 2008). Note: error bars represent the standard deviation of the total concentration.**

composition. For example, Laurent et al. (2009) reported that smaller floc size was critical for increased Cd sorption. Activated sludge (of similar dry weight) comprised of smaller flocs has better availability of binding sites due to increased floc surface area. In this study median floc sizes reduced from 274 to 164 µm between SRTs of 10 and 27 days (Table S2). This may explain improved removal achieved by the 27 day SRT biomass. However, metal concentrations in the aqueous phase of wastewater (to which EQS's are applied) increased during treatment at all SRTs (Table 1). Aqueous removals of Cu and Zn ranged from -34 to -91% at 3, 10 and 27 day SRTs. Increased aqueous metal concentration is characteristic of ASP treatment and aqueous removals of -170% have previously been observed for Zn at an SRT of 8 days (Santos et al., 2010).

Lengthening HRT of the pilot plant system initially resulted in increased removal of  $\sum_{\text{METAL}}$ 's observed during primary sedimentation (prior to secondary treatment). This was a result of a high proportion of metals found in crude sewage being associated with particulates. For example, the percentage of Zn and Cu within the particulate phase (>0.45 µm) of crude wastewater was  $88 \pm 4\%$  and  $97 \pm 2\%$ , respectively (Fig. 4). Suspended solids concentration of crude sewage was  $366 \pm 154 \text{ mg l}^{-1}$  and reduced to  $123 \pm 15$ ,  $88 \pm 7$  and  $64 \pm 13 \text{ mg l}^{-1}$  in settled sewage for primary sedimentation times of 4, 8 and 12 h respectively (Table S3). This

corresponded well to metal behaviour. To demonstrate,  $\sum_{\text{METAL}}$  concentrations in crude sewage were  $141 \pm 29 \text{ µg l}^{-1}$  with the majority of this attributed to Zn ( $91 \pm 27 \text{ µg l}^{-1}$ ) and Cu ( $46 \pm 11 \text{ µg l}^{-1}$ ) (Fig. 4). In settled sewage Zn concentrations were  $79 \pm 18$ ,  $41 \pm 6$  and  $26 \pm 5 \text{ µg l}^{-1}$  corresponding to primary sedimentation residence times of 4, 8 and 12 h (Table 1). Similarly, total Cu concentrations were reduced to  $39 \pm 8$ ,  $27 \pm 4$  and  $17 \pm 2 \text{ µg l}^{-1}$  in settled sewage. Metal concentrations found in settled sewage were therefore reduced as HRT of the pilot plant was lengthened.

During secondary treatment, removal of  $\sum_{\text{METAL}}$ 's decreased with longer HRT. This is explained by a linear correlation ( $r^2$  0.92) with reduced  $\sum_{\text{METAL}}$  concentrations in settled sewage (as primary sedimentation residence time increased) (Fig. 5). Secondary treatment at longer HRT also resulted in increased production of aqueous metals (Fig. 5). To illustrate, aqueous metals removals were  $-88 \pm 9\%$  at an 8 h HRT whereas removals at 16 and 24 h HRTs were  $-176 \pm 9\%$  and  $-128 \pm 6\%$  respectively. Individually, Cu exhibited the greatest negative removal. Secondary removals were  $-40 \pm 12\%$  (8 h HRT),  $-253 \pm 53\%$  (16 h HRT) and  $-190 \pm 24\%$  (24 h HRT). It is postulated that longer contact times may facilitate further oxidation of particulate organic matter resulting in the increased solubilisation of metals (Ziolko et al., 2011). Also, products of bacterial metabolism and the production of extracellular polymers sloughed from bacterial

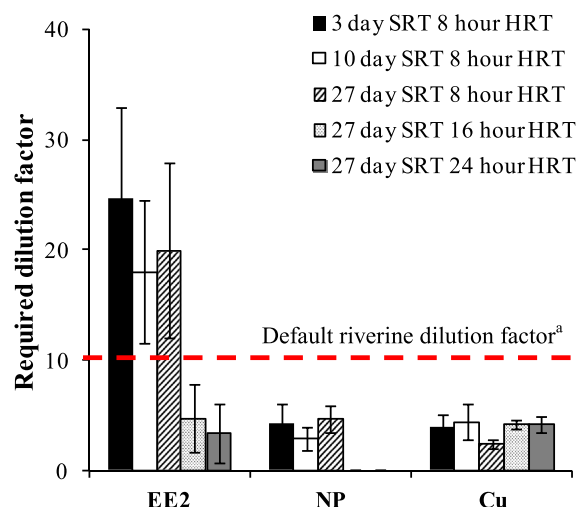


**Fig. 5 – Impact of  $\Sigma_{\text{METAL}}$  concentration (i.e., sum of aqueous and particulate concentrations of Zn, Cu, Pb, Cd and Ni) in settled sewage to secondary (total and aqueous metals) removal.**

cells could help retain metals in the bulk solution at longer HRTs (Santos et al., 2010). This increased solubilisation of metals at longer HRT is significant as only the aqueous (i.e., pre-filtered) phase of water matrices is to be considered for EQS compliance (European Commission, 2008).

### 3.2. Final effluent quality and the prospect of achieving environmental compliance

To assess the likelihood of achieving compliance for each micropollutant within the receiving aqueous environment, the level of final effluent dilution required for consent was determined (Fig. 6). This denotes the minimum river dilution requirement to achieve the EQS or proposed legislative target for each ASP final effluent for EE2, NP and Cu (only aqueous concentrations are considered for compliance). These chemicals were selected as they required the most dilution in this study within the micropollutant groups; steroid estrogens, NPx's and metals. Compliance was unequivocally assured for a default dilution factor of 10 (European Commission, 2003) by the 27 day SRT 16 and 24 h HRT operations. Modifying ASP operation had the greatest impact to the dilution requirement for EE2. This was due to EE2 having a very low proposed legislative target of  $3.5 \times 10^{-5} \mu\text{g l}^{-1}$  (European Commission, 2012; Table S1) and it being more responsive to changes in HRT. Dilution of  $\geq 20$  times was needed to ensure consent for ASPs where SRT was investigated (Fig. 6). Conversely, at 16 and 24 h HRTs the dilution required was  $\leq 10$  due to increased removals of EE2. In comparison, the required dilution for consent for NP was  $< 10$  for all ASP final effluents. However, despite NP being the only chemical within the NPx group which has an EQS, it has been successfully demonstrated that the biotransformation of readily degradable NP precursors in the environment (which are not legislated for) could theoretically lead to a breach of EQS downstream of the effluent discharge point (Petrie et al., 2013b). Molar concentrations of cumulative NPx's in final effluents were observed up to  $37 \pm 6 \mu\text{mol m}^{-3}$



**Fig. 6 – Riverine dilution required to ensure consent to EQS and proposed legislative targets for EE2 (proposed legislative target =  $3.5 \times 10^{-5} \mu\text{g l}^{-1}$ , NP (EQS =  $0.3 \mu\text{g l}^{-1}$ ) and Cu (proposed legislative target =  $1\text{--}28 \mu\text{g l}^{-1}$ ) at each ASP operation. <sup>a</sup>Default riverine dilution factor of 10 as described by the European Commission (European Commission, 2003) Note: Dilution factor required for consent is calculated from the aqueous (i.e., pre-filtered) concentration only (European Commission, 2008). Micropollutant concentration already present in dilution water is assumed to be zero. For metals, a calcium carbonate concentration of  $0\text{--}50 \text{mg l}^{-1}$  is assumed as EQSs are dependent on water hardness.**

( $1.4 \mu\text{mol m}^{-3}$  of NP is equivalent to its EQS of  $0.3 \mu\text{g l}^{-1}$ ). It is therefore advantageous to operate the ASP at conditions which enhance biotransformation across the NPx family of chemicals (i.e., high SRT and long HRT conditions).

Interestingly, aqueous metal concentrations (to which EQS's are applied) were similar to, or below their proposed legislative targets for consent in crude and settled sewages (Fig. 4). Following aerobic treatment, aqueous concentrations were above these targets for both Cu and Zn. The 27 day SRT 8 h HRT achieved the best final effluent quality with respect to aqueous Cu concentrations. The required dilution factor needed to achieve the proposed legislative target for consent was  $2.4 \pm 0.4$  times (Fig. 6). Despite lower settled sewage concentrations at longer HRTs due to improved removal of particulate bound metals during primary sedimentation, no improvement in the required dilution factor for consent was observed. This is because increased solubilisation of the remaining particulate bound phase was observed. However, the dilution requirement is likely to be greater for a full-scale process operating at similar HRT conditions. This is because primary sedimentation tanks are not designed for the longer residence times observed in this study and therefore, settled sewage metal concentrations are likely to be considerably greater. For example, influent wastewaters of full-scale processes commonly contain Cu and Zn at concentrations  $> 100 \mu\text{g l}^{-1}$  (Rule et al., 2006; Ziolkko et al., 2009; Gardner et al., 2013). Consequently, poorer secondary removal of aqueous

metals at longer HRT will be more apparent in final effluents here. This increase of aqueous metal concentrations at conditions favourable for organic micropollutant removal demonstrates the difficulty of optimising ASP operation for simultaneous removal of estrogens, NPx and metals.

### 3.3. Options for upgrading existing ASPs for simultaneous micropollutant removal

The level of micropollutant removal required to reach compliance limits for a given ASP will be highly site specific. This will depend on receiving micropollutant concentrations as well as available dilution at the discharge point. Despite the very different behaviour observed for these micropollutants during ASP treatment, operating conditions can be modified to improve their removal simultaneously. The majority of existing full-scale ASPs are designed to operate at mid-ranged conditions (i.e., circa 10 day SRT 8 h HRT) at yearly average flows (Aboobakar et al., 2013). Yet these tend to receive significant variations in sewage flow resulting in a dynamic system with significant variations in SRT and HRT. Therefore improvements in full-scale performance are achievable by better process control to improve process continuity and reduce variations in both SRT and HRT. For example, the use of *in-situ* suspended solids probes and real-time flow measurements could help avoid variability in SRT. It was demonstrated that operation at a higher SRT improved simultaneous micropollutant removal. However, higher SRT operation has traditionally been associated with significantly greater aeration demands. Recently it has been shown that operation at a higher SRT achieves improved oxygen transfer due to smaller activated sludge floc size and better uniformity (Leu et al., 2012). Consequently, aeration requirements are not as great as previously considered.

Achieving a significant improvement to HRT will likely require infrastructure development. Where sufficient space is not available to achieve a desired HRT, continuous use of onsite storm tanks or remote holding tanks could act as a buffer to counter-balance fluctuations in sewage flow, enabling process continuity whilst operation at a longer HRT. Furthermore, lengthening secondary HRT is likely to require improved metals removal during primary treatment as they behaved very differently to organics during biological treatment. Therefore, maximising particulate bound metals removal during primary sedimentation to circumvent their solubilisation during secondary treatment is essential. This is critical for sites which have low dilution ratios or where the receiving concentrations of metals are comparatively greater. This study successfully demonstrated that primary sedimentation tanks operated at longer HRT can effectively enhance metal removal prior to secondary treatment (Fig. 4). However, enhancing suspended solids removal by existing assets may not be achievable without the continual dosing of coagulants. For example, there may not be sufficient space available onsite to increase primary sedimentation tank size to facilitate HRTs similar to those utilised here. As a trade-off, the alternative or complimentary use of micro-screens can yield improved suspended solids removal at a comparatively smaller footprint (Salsnes Filter, 2011).

## 4. Conclusions

- Prospects of achieving EQS compliance through ASP operation improvements are highly site specific, governed by receiving micropollutant concentrations in crude sewage, available dilution water and quality, and available onsite space for infrastructure development.
- Solids retention times of 10 and 27 days achieved greater  $\sum_{EST}$  removal and an increase of SRT from 10 to 27 days achieved greater  $\sum_{NPx}$  and  $\sum_{METAL}$  removal.
- Longer HRT of the aerobic treatment stage was vital for achieving increased biodegradation of  $\sum_{EST}$  (specifically EE2) and suitable precursor biotransformation such that positive NP removals are achieved.
- Enhanced removal of particulates from crude wastewater during primary sedimentation is critical for improved metal removals and avoiding increased concentrations of aqueous metals following secondary treatment at longer HRT.
- This study has identified that the broad range of micropollutant chemistries do result in trade-offs which should be considered for future studies.

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## Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.watres.2014.05.036>.

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