



Characterisation of groundwater dissolved organic matter using LC–OCD: Implications for water treatment

Helen Rutlidge^{a,b,*}, Liza K. McDonough^{a,c}, Phetdala Oudone^{a,c}, Martin S. Andersen^{a,b}, Karina Meredith^d, Khorshed Chinu^e, Mark Peterson^d, Andy Baker^{a,c}

^a Connected Waters Initiative Research Centre, UNSW Sydney, NSW, 2052, Australia

^b School of Civil and Environmental Engineering, UNSW Sydney, NSW, 2052, Australia

^c School of Biological, Earth and Environmental Sciences, UNSW Sydney, NSW, 2052, Australia

^d Australian Nuclear Science and Technology Organisation (ANSTO), New Illawarra Rd, Lucas Heights, NSW, 2234, Australia

^e Mark Wainwright Analytical Centre, UNSW, Sydney, NSW 2052, Sydney, Australia

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ABSTRACT

The polarity and molecular weight of dissolved organic matter (DOM) is an important factor determining the treatability of water for domestic supply. DOM in surface water and groundwater is comprised of a mixture of carbon with varying molecular weight ranges, with its composition driven by DOM sources and processing. Here, we present the largest dataset of chromatographic DOM in surface and groundwater samples ($n = 246$) using liquid chromatography organic carbon detection (LC–OCD). Our data represents four categories (surface water, hyporheic zone water, local groundwater, and regional groundwater) from five different sites across Australia. In all environments, high molecular weight hydrophilic DOM such as biopolymers (BP) and humic substances (HS) are present in surface waters and are processed out of groundwater as it moves from surface water and hyporheic zones into shallow local groundwater and deeper regional groundwaters. This results in a higher percentage of low molecular weight neutrals (LMWN) and hydrophobic organic carbon (HOC) in deeper regional groundwaters. Our findings indicate that the presence of sedimentary organic matter strongly influence the character of surface and groundwater DOM, resulting in groundwater with higher HS aromaticity and molecular weight, and reduced percentage of LMWNs. We also observe highly variable hydrophilic / HOC ratios in groundwater at all sites, with 9.60% and 25.64% of samples at sites containing sedimentary peat layers and non-sedimentary peat sites respectively containing only hydrophilic dissolved organic carbon (DOC). We identify average hydrophilic / HOC ratios of 4.35 ± 3.76 and 7.53 ± 5.32 at sites containing sedimentary peat layers and non-sedimentary peat sites respectively where both hydrophilic DOC and HOC are present. Overall our results suggest that fractured rock and alluvial aquifers in sedimentary organic carbon poor environments may contain DOC which is better suited to ozonation, biologically activated carbon filtration powdered activated carbon, suspended ion exchange treatment or magnetic ion exchange resin since DOC is more hydrophilic and of lower molecular weight and lower aromaticity. Aquifers located near sedimentary organic matter layers may benefit from pre-treatment by coagulation/flocculation, sedimentation and sand filtration which have high removal efficiency for high molecular weight and polar compounds.

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1. Introduction

Groundwater is the world's largest active source of fresh water and is heavily relied on for agricultural irrigation, industrial uses and drinking water supply. Currently, 98.5% of the total domestic self-supplied fresh water in the United States is obtained from

groundwater, with some states including Hawaii, Florida, Idaho and Mississippi relying on groundwater for 87 - 95% of their total public water supply (Dieter et al., 2018). Understanding dissolved organic matter (DOM) concentration and character in groundwater is important, as this ultimately determines its treatability for domestic water supply (Regan et al., 2017). DOM is comprised of a complex mixture of high and low molecular weight fractions. The relative proportions of these fractions in natural waters is determined by both its source and the type and extent of processing it has undergone (Amon and Benner 1996). Presently, there are

* Corresponding author.

E-mail addresses: h.rutlidge@unsw.edu.au (H. Rutlidge), l.mcdonough@unsw.edu.au (L.K. McDonough).

minimal studies regarding the character of DOM in natural groundwater systems in various environmental settings. These are vital to understand if we are to optimise the use of groundwater for domestic water supply.

Processes including sorption, photodegradation and microbial processing (Amon and Benner 1996; Chapelle et al., 2013; Shen et al., 2015; Singh et al., 2016) fractionate DOM of different molecular weights and polarity, altering the composition of DOM remaining in solution. For example, high molecular weight (HMW) and vascular plant derived aromatic DOM are more susceptible to sorption, whilst carboxyl-rich aliphatics and lower molecular weight DOM compounds are preferentially retained in solution (Lv et al., 2016; Riedel et al., 2016). Hydrophobic DOM has also been shown to be preferentially adsorbed over hydrophilic DOM to soils and hydrous oxides, with a displacement of hydrophilic DOM by hydrophobic DOM possible in environments with limited surface binding sites (Kaiser and Zech 1997).

Bertilsson and Tranvik (2000) identified a strong correlation between level of UV exposure and dissolved organic carbon (DOC) concentration of samples obtained from 38 lakes ($R^2 > 0.80$). Aromatic DOM groups are particularly susceptible to photodegradation (Jardine et al., 1989; Kaiser et al., 1996; Kattner et al., 2006; Stubbins et al., 2010). In contrast, lower molecular weight and aliphatic molecules are more photo-resistant and can even be produced by photo-degradation of other DOM compounds (Bittar et al., 2015; Riedel et al., 2016; Stubbins et al., 2010). Where environmental conditions permit a significant amount of DOM photodegradation in surface waters, aquifers recharged by these waters may contain higher amounts of low molecular weight DOM compared to aquifers recharged by rainfall through soils or impacted by sedimentary organic carbon which has been protected from photodegradation.

Here, we aim to characterise groundwater DOM using a size exclusion technique known as liquid chromatography organic carbon detection (LC-OCD) in five sites across Australia and use these data to infer suitability of groundwater DOM for water treatment and domestic water supply. Bagoth et al. (2011) used LC-OCD to demonstrate the removal of the biopolymer fraction (from 10 to 5%) during the coagulation of river source waters, and the addition of building blocks (from 17% to 22%) from the treatment process. Andersson et al. (2020) compared LC-OCD data from a pilot plant using suspended ion exchange (SIX) treatment and a full-scale works using aluminium sulfate and sand filtration. LC-OCD demonstrated that the treated lake water differed in LC-OCD properties, with coagulation removing the biopolymers and higher-MW HS fraction, and SIX was less selective and removed relatively more of the medium-MW range HS fraction. In a series of jar-test experiments, Shutova et al. (2020) investigated the role of powdered activated carbon (PAC) in water treatment at two surface water reservoirs. LC-OCD results show that with the treatment by the coagulation-flocculation-sedimentation, DOM removal efficiency was 65% with biopolymer and HS fractions preferentially removed. Adding PAC to the coagulation-flocculation-sedimentation process greatly improved the removal of all DOM fractions identified by LC-OCD. We hypothesise that sample depth will play a role in determining the character of DOC due to processing mechanisms in the subsurface, with high molecular weight DOM likely processed as it moves through groundwater systems and into deeper regional groundwaters. We also hypothesise that the presence of sedimentary peat layers in the subsurface in some locations may also impact groundwater DOM character due to potentially large inputs of high molecular weight and unprocessed DOC into groundwaters at these sites. This study represents the largest characterisation of chromatographic DOM in groundwater to date, representing a variety of aquifer depths (1.90 - 40.7 m), hydrological environment and geological environments.

2. Materials and methods

2.1. Sampling sites

Four regions around New South Wales (NSW) and one in Western Australia (WA), Australia were sampled for groundwater, hyporheic and surface water DOC, as conditions allowed. The sampling locations include Maules Creek, Anna Bay, Wellington and Thirlmere in NSW, and Rottneest Island in WA. The locations are shown in Fig. 1.

Two of the sampling sites, Thirlmere Lakes and Anna Bay are associated with lentic water bodies and contain sedimentary peat layers. Samples at Anna Bay were taken from Samuray Beach near a wetland behind unstable calcareous sand dunes (Bell 1997) and from a transect of bores positioned at various depths between the wetland and the coastline (McDonough et al., 2020a; Meredith et al., 2020). This aquifer is part of the Tomaree Groundwater Source that is used for potable water supply (Meredith et al., 2020). Thirlmere Lakes are located in an uplifted entrenched meander (Timms 1992) approximately 37 km from the eastern Australian coastline. There is no surface water flow between the lakes and substantial peat layers have formed in the lake beds. Local groundwater samples from this site were obtained from pre-existing bores located adjacent to Lake Couridjah, Lake Gandangarra and Lake Nerrigorang. Hyporheic samples were collected from piezometers installed adjacent to Lake Baraba, Lake Couridjah and Lake Werri Berri.

Maules Creek, Wellington and Rottneest Island represent alluvial, fractured rock and karst environments and are referred to as the non-sedimentary peat sites. The sites of Wellington and Maules Creek are both associated with intermittent streams (lotic systems) and located in semi-arid inland sites between approximately 250 - 270 km from the eastern Australian coast. The Maules Creek Catchment is comprised of Quaternary alluvial sediments overlying Permian volcanics, consolidated sedimentary rocks and coal measures (Andersen and Acworth 2009; Giambastiani et al., 2012; Kelly et al., 2013; McCallum et al., 2013). Samples were collected at Maules Creek from pre-existing bores located in transects from Maules Creek, Horsearm Creek and Middle Creek (Cuthbert et al., 2016; Hartland et al., 2015; Rau et al., 2017). Samples from Wellington were collected from pre-existing bores located in alluvial sediments near river systems (Graham et al., 2015a; Keshavarzi et al., 2017), fractured metasediments (Graham et al., 2015b) as well as from thinly bedded and massive Devonian limestone (Strusz 1965) at the Wellington Caves Complex. At Wellington groundwater is abstracted for local stock and domestic supply and some local irrigation (Keshavarzi et al., 2017).

Rottneest Island is located approximately 18 km off the south west coast of Western Australia (Fig. 1). Because of its highly permeable carbonate aeolianite deposits the island has no fresh surface water features. Groundwater samples were collected from the carbonate aeolianite aquifer (Bryan et al., 2020) which was formed in the early Holocene (Price et al., 2001). This aquifer has been used for some of the island's potable water supply during the period 1977 - 2017 (Bryan 2017). This site features a Mediterranean climate type with hot, dry summers and mild, wet winters (Bryan et al., 2016).

2.2. Sample collection and analysis

The five sampling sites were sampled at different times predominantly between 2016 and 2019 with earlier trips for both Anna Bay and Rottneest Island. Details of the sampling trips are given in Table 1. During each trip, sampling was performed to collect for DOC and LC-OCD analysis. This dataset represents 246

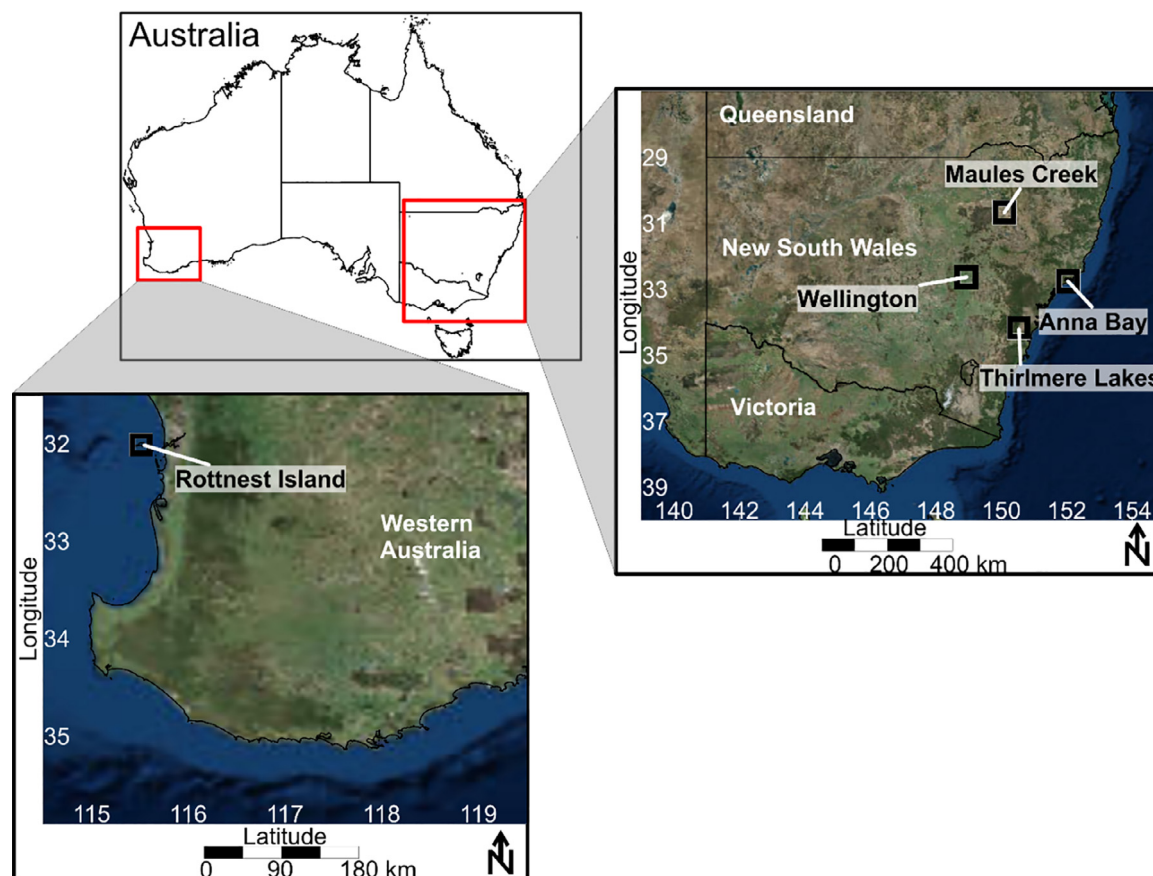


Fig. 1. Sampling locations in NSW and WA, Australia.

samples in total representing 131 unique locations with 8 surface water bodies, 45 hyporheic, 71 local groundwater and 7 regional groundwater locations.

Prior to sampling the groundwater samples, a dip metre was used to record standing water levels. An in-line Sheffield flow-cell attached to HACH HQ40D multimeters measuring electrical conductivity (EC), pH, temperature, and dissolved oxygen (DO) were used to observe purging. Once parameters stabilised, samples were then collected. Surface water samples and hyporheic samples from Maules Creek were obtained using a Series II Geopump peristaltic pump and sampling spear (10 mm diameter, 50 mm long screen). Hyporheic samples from Thirlmere Lakes were obtained using a Series II Geopump peristaltic pump from 50 mm diameter PVC piezometers with screen lengths between 0.4 and 1.4 m. Groundwater samples were obtained using a Monsoon 12-volt groundwater pump for sampling 50 mm diameter PVC piezometers with screen lengths between 0.5 and 1 m.

LC-OCD and total DOC samples were collected in 60 mL Falcon™ tubes with no headspace after filtering in-field at 0.45 µm using Waterra polyethersulphone in-line filters and analysed using a DOC-LABOUR LC-OCD size-exclusion chromatography system and a total organic carbon (TOC) analyser (Aurora 1030 wet oxidation TOC analyser, OI Analytical, College Station, Texas, United States) respectively at University of New South Wales, Sydney (UNSW). LC-OCD utilizes a size exclusion chromatography technique to determine DOM concentration and assign DOM into major fractions based on molecular weight (Huber et al., 2011). LC-OCD is based on the principle that different sizes of organic matter molecules have different retention times on LC-OCD column material. Larger

molecules have shorter elution times as they are unable to diffuse into column material, whilst smaller molecules diffuse, leading to longer retention times. The result of a LC-OCD analysis is a chromatogram of peaks based on retention time and signal intensity, which the user must then manually process in order to quantify and distinguish between different DOM molecular size fractions. The International Humic Substance Society (IHSS) Suwannee River humic and fulvic acid standards are regularly used every 3–4 months to provide the basis of the retention times that are used as part of this process. According to Huber et al. (2011) the fractions are referenced as biopolymers (BP, molecular weight > 20,000 Da), humic substances (HS, 500 Da < molecular weight > 10,000 Da), building blocks (BB, 300 Da < molecular weight > 500 Da), low molecular weight acids (LMWA, molecular weight < 350 Da), low molecular weight neutrals (LMWN, < 350 Da) and hydrophobic organic carbon (HOC), which is classified as the material which does not leave the column. The sum of the BP, HS, BB, LMWN and LMWA fractions represent the hydrophilic organic carbon (CDOC). These fractions are labelled throughout this manuscript to remain consistent with the fractions defined by Huber et al. (2011). We do however note that each fraction is defined only by the molecular weight and hydrophobicity of the molecules, therefore no information on the molecular composition or origin of the molecule should be inferred by these groupings. For example, molecules defined as HS may therefore not necessarily represent a humic origin, and thus caution should be taken with the interpretation of these data. Due to high DOC concentrations, some samples were diluted with ultra-pure water prior to analysis with a maximum dilution factor of 6.

Table 1
Sampling details for each site.

Site	Date	Samples
Anna Bay	February 2014	15 local groundwater
	February 2018	21 hyporheic
		10 local groundwater
	March 2018	17 hyporheic
	March 2018	18 hyporheic
	April 2018	17 hyporheic
Maules Creek	May 2018	17 hyporheic
		3 local groundwater
	February 2016	3 surface water
		16 hyporheic
		6 local groundwater
		5 regional groundwater
Rottnest Island	October 2017	1 surface water
		5 local groundwater
		2 regional groundwater
	March 2018	1 surface water
		5 local groundwater
		2 regional groundwater
Thirlmere Lakes	September 2014	20 local groundwater
	March 2015	7 old seawater
		1 surface water
	March 2017	2 local groundwater
		1 surface water
		6 local groundwater
Wellington	March 2019	2 surface water
		5 hyporheic
	May 2019	1 surface water
		4 hyporheic
	August 2019	2 surface water
		5 hyporheic
Wellington	April 2017	3 local groundwater
	August 2017	8 local groundwater
		2 surface water
		5 local groundwater
	March 2019	1 surface water
		7 local groundwater

2.3. Statistical analysis

One-way ANOVA and Tukey's post-hoc test in RStudio was used to test if there were significant differences for the different LC-OCD fractions between the sites, in addition to differences due to sample class (surface water, hyporheic zone and local and regional groundwater). To test which fractions displayed differences between the sites the local groundwater samples from each site were used for the ANOVA analysis as this was the only sample class present across all sites. For the depth comparison, ANOVA was performed separately for each individual site for the fractions that were shown to have significant differences between the sites,

while for the other fractions ANOVA was performed on the whole dataset. Redundancy analysis (RDA) was used as a constrained ordination approach to compare the presence of sedimentary organic matter, and sample depth categories with LC-OCD fractions. RDA was conducted using the vegan package in RStudio (v. 1.1.456), with LC-OCD fractions scaled to their unit variances.

3. Results

The total DOC for the majority of the samples were analysed by two techniques, the Aurora TOC analyser and LC-OCD. The linear equation for the comparison between the TOC analyser and LC-OCD for total DOC for samples analysed by both techniques has a slope of 0.99, an intercept of -0.22 and an R^2 of 0.95 (Figure S1).

3.1. Site comparison of DOC and LC-OCD fractions

The average concentration of DOC in groundwater and surface water for all samples was $4.29 \text{ mg/L} \pm 4.83$ (1σ) and $10.78 \text{ mg/L} \pm 15.89$ (1σ) respectively. The average concentration in the hyporheic zone (depths of $> 0 \text{ m} < 3 \text{ m}$ bgs), local groundwater (depths of $> 3 \text{ m}$ bgs) and regional groundwater was $5.66 \text{ mg/L} \pm 5.24$, $3.02 \text{ mg/L} \pm 3.90$ and $0.51 \text{ mg/L} \pm 0.12$ respectively. The average concentration for each sample type at each site is given in Table 2. The Thirlmere Lakes surface water and hyporheic samples contained significantly higher concentration of total DOC and individual fractions. The DOC concentration was lower for the groundwater samples compared to the surface and hyporheic samples for three out of the five sites with the exceptions of Anna Bay (due to peat being present from 5 m below ground level) and Rottnest Island. LMWA were only detected in Rottnest Island and Thirlmere Lakes samples.

All sites include local groundwater samples. A comparison of the average composition (based on the percentage of each fraction) of the local groundwater samples at each site is shown in Fig. 2 and the ANOVA results in Table 3. For all sites, except Thirlmere Lakes, the HS fraction was dominant, with BP the least abundant. The relative abundance of HOC, HS, and LMW-N fractions were significantly different between the sites, whilst BP and BB were similar.

3.2. Changes in groundwater DOC with depth and the presence of peat layers

The ANOVA results (Table 3) and Fig. 2 indicate that the presence of sedimentary peat layers plays a part in the local groundwater differences between the sites in terms of the fractions. The

Table 2
Average concentrations (mg / L) of DOC and LC-OCD fractions at each site.

Site	Type	DOC $\pm 1\sigma$ (mg/L)	HOC $\pm 1\sigma$ (mg/L)	BP $\pm 1\sigma$ (mg/L)	HS $\pm 1\sigma$ (mg/L)	BB $\pm 1\sigma$ (mg/L)	LMWN $\pm 1\sigma$ (mg/L)	LMWA $\pm 1\sigma$ (mg/L)
Anna Bay	Hyporheic	4.72 ± 2.48	0.79 ± 0.76	0.04 ± 0.06	2.58 ± 1.40	0.65 ± 0.39	0.67 ± 0.32	0.00 ± 0.00
	Local GW	7.31 ± 5.33	1.60 ± 1.39	0.02 ± 0.03	3.99 ± 3.03	1.02 ± 0.73	0.68 ± 0.46	0.00 ± 0.00
Maules Creek	Surface (river)	1.87 ± 0.87	0.11 ± 0.10	0.18 ± 0.12	0.99 ± 0.51	0.13 ± 0.07	0.46 ± 0.29	0.00 ± 0.00
	Hyporheic	1.83 ± 1.54	0.47 ± 1.19	0.01 ± 0.01	0.79 ± 0.34	0.13 ± 0.10	0.43 ± 0.12	0.00 ± 0.00
	Local GW	1.02 ± 0.29	0.17 ± 0.12	0.01 ± 0.01	0.48 ± 0.19	0.13 ± 0.07	0.25 ± 0.07	0.00 ± 0.00
	Regional GW	0.51 ± 0.12	0.14 ± 0.08	0.00 ± 0.00	0.10 ± 0.03	0.07 ± 0.06	0.20 ± 0.10	0.00 ± 0.00
Rottnest Island	Surface (ocean)	0.98 ± 0.30	0.00 ± 0.00	0.12 ± 0.03	0.51 ± 0.16	0.08 ± 0.02	0.26 ± 0.14	0.01 ± 0.01
	Local GW	1.56 ± 0.81	0.21 ± 0.11	0.00 ± 0.00	0.87 ± 0.55	0.22 ± 0.11	0.26 ± 0.14	0.02 ± 0.01
	Old seawater	1.91 ± 0.81	0.05 ± 0.13	0.00 ± 0.00	1.27 ± 0.55	0.27 ± 0.19	0.28 ± 0.14	0.04 ± 0.01
Thirlmere Lakes	Surface (lake)	28.3 ± 17.08	5.51 ± 5.87	2.31 ± 0.85	10.90 ± 5.78	4.21 ± 2.54	4.08 ± 1.47	1.32 ± 1.74
	Hyporheic	16.0 ± 7.94	2.39 ± 1.70	0.80 ± 0.81	7.78 ± 4.60	2.53 ± 1.69	2.27 ± 0.76	0.60 ± 0.62
	Local GW	3.24	1.41	0.01	0.60	0.26	0.96	0.00 ± 0.00
	Regional GW	0.35 ± 0.07	0.16 ± 0.04	0.00 ± 0.00	0.05 ± 0.01	0.04 ± 0.01	0.09 ± 0.00	0.00 ± 0.00
Wellington	Surface (river)	6.36 ± 3.26	0.21 ± 0.13	0.26 ± 0.18	3.98 ± 2.07	1.09 ± 0.59	0.81 ± 0.38	0.00 ± 0.00
	Local GW	1.27 ± 0.57	0.11 ± 0.05	0.01 ± 0.00	0.70 ± 0.36	0.18 ± 0.16	0.28 ± 0.11	0.00 ± 0.00

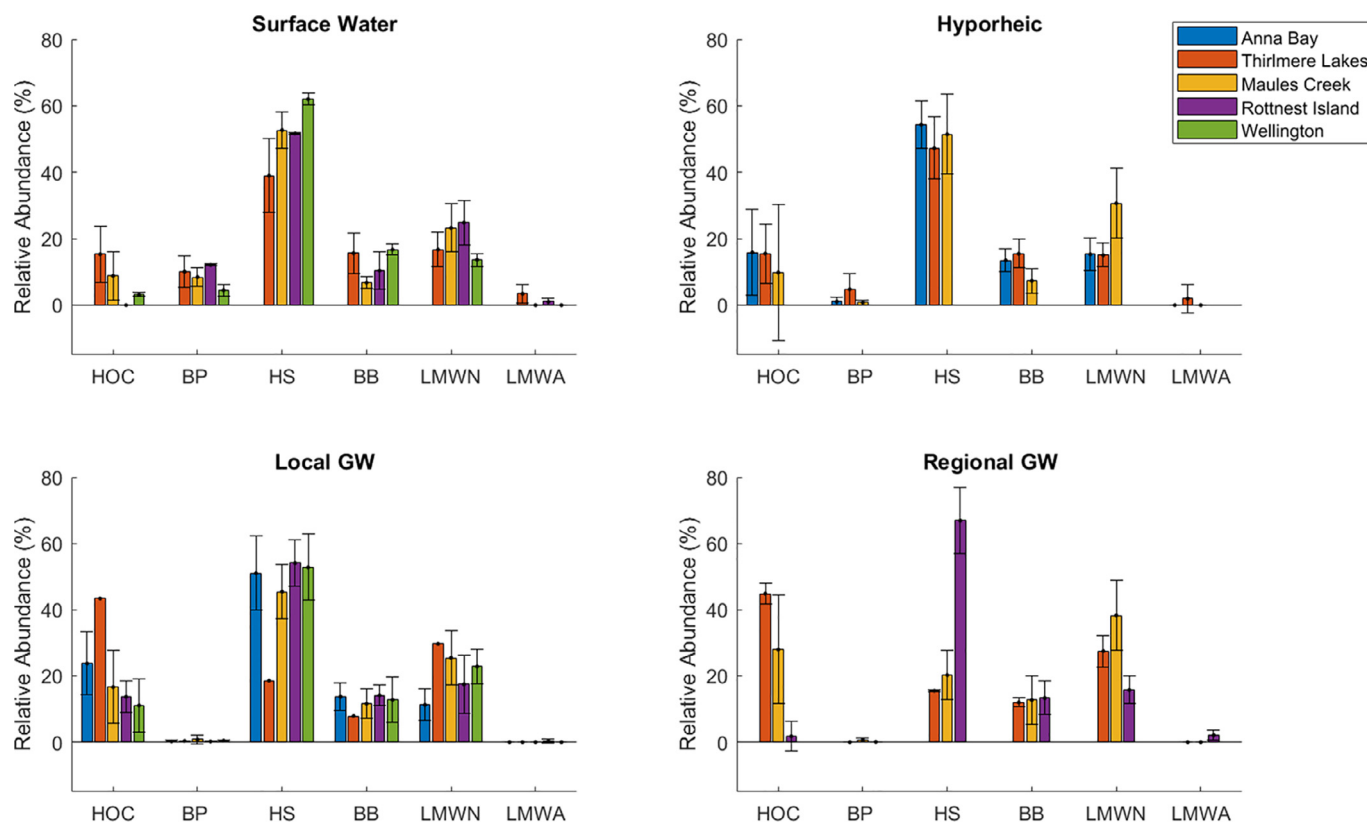


Fig. 2. The DOC relative composition for each depth category at each site with the error bars representing $\pm 1\sigma$.

Table 3

The ANOVA results (p-values) for the site comparison using the local groundwater samples. p-values in bold indicate significant difference between the sites with a 95% confidence interval.

	p-value
DOC (mg/L)	$<2 \times 10^{-16}$
HOC (mg/L)	3.1×10^{-12}
BP (mg/L)	$<2 \times 10^{-16}$
HS (mg/L)	$<2 \times 10^{-16}$
BB (mg/L)	$<2 \times 10^{-16}$
LMWN (mg/L)	$<2 \times 10^{-16}$
HOC (%)	1.6×10^{-11}
BP (%)	0.16
HS (%)	5.1×10^{-10}
BB (%)	0.29
LMWN (%)	1.9×10^{-9}

sites that contained sedimentary peat layers (i.e. Anna Bay and Thirlmere Lakes) were higher in HOC (%) (significant at Thirlmere Lakes) compared to the alluvium and karst sites (i.e. Maules Creek, Wellington and Rottnest Island). Of the alluvium and karst sites, Maules Creek has the highest amount of HOC (%), which we hypothesise is due to the presence of aged organics at depth at this site (McDonough et al. 2020b). While for HS (%), Thirlmere Lakes was significantly lower than the other sites and with LMWN (%) Anna Bay was significantly lower than the other sites. Overall, the two sites with sedimentary peat layers contained a higher average concentration of all LC–OCD fractions for the equivalent depth than the other sites (Table 2).

All sites, over the course of sampling, appeared to have surface water systems infiltrating the aquifers (i.e. losing conditions) (see supplementary material for details) which allows us to in-

fer changes in DOC composition along a flow path by looking at the changes in proportion (the relative abundance of each fraction expressed as percentage of total DOC) of the different fractions from surface water to hyporheic to local groundwater to regional groundwater (Fig. 3). The ANOVA results for this depth comparison are given in Table 4. Moving along this inferred flow path, HOC (%) showed an increase (significant at all sites except for Rottnest Island) and the HS (%) showed a decrease (significant at the Maules and Thirlmere sites). The LMWN (%) fraction also had significant differences between the different depths for all sites, with general increase with depth. For all sites, the percent of BP are significantly higher in the surface water samples than the subsurface samples. Fig. 3 shows that the BP fraction is almost entirely removed at all sites from surface to hyporheic zone water. In general, there is a decline the concentration of the various fractions from surface water through to hyporheic zone to local and regional groundwater. Surface water HS molecular weight and aromaticity show very similar average values for both sedimentary peat and non-sedimentary peat sites (Fig. 3), however, the presence of peat layers in the subsurface appears to result in an increase in HS molecular weight and aromaticity in the local groundwater samples.

RDA analysis of the dataset also show that DOC concentration and composition differs for peat and non-peat sites (Fig. 4A). The first axis in Fig. 4A (RDA1) describes the presence or absence of sedimentary peat layers. Non-sedimentary peat sites have negative values on RDA1, whilst sites with sedimentary peat layers have the positive value on RDA1. Positive values on RDA1 represent high HS (%), high HS aromaticity, high DOC concentrations and high HS molecular weight, whilst negative values on RDA1 represent high LMWN (%) and BB (%), though the abundance of BB fractions are very low and hence would have little influence. The second axis (RDA2) on Fig. 4A indicates that DOC hydrophobicity is related to

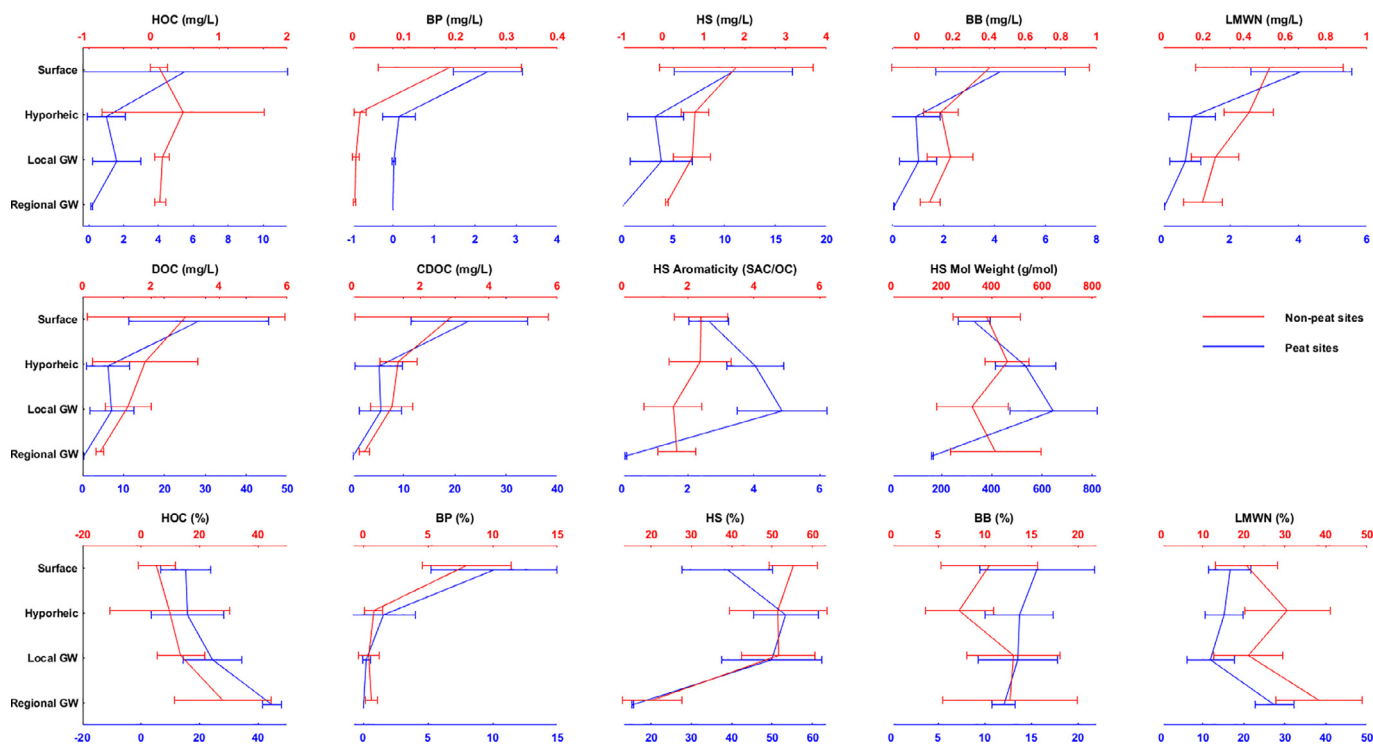


Fig. 3. Concentrations of DOC and hydrophilics, humic substances (HS) aromaticity (calculated as per Huber et al. (2011) and defined as spectral absorption coefficient/organic carbon concentration (SAC/OC)), HS molecular weight (in g/mol) and concentrations (mg/L) and relative abundance (expressed as percent of total DOC) of biopolymers (BP), building blocks (BB), low molecular weight neutrals (LMWN), HS and hydrophobic organic carbon (HOC) with depth for the samples containing sedimentary peat layers ($n = 140$) and non-sedimentary peat ($n = 107$) samples. Error bars represent 1 standard deviation.

Table 4

The ANOVA results (p-values) for the depth comparison. p-values in bold indicate significant difference between the different depths with a 95% confidence interval.

	Sites combined	Anna Bay	Thirlmere Lakes	Maules Creek	Rottnest Island	Wellington
HOC (%)		0.0037	0.0002	5.3×10^{-5}	0.019	0.12
BP (%)	$<2 \times 10^{-16}$					
HS (%)		0.47	0.00041	6.3×10^{-12}	0.49	0.14
BB (%)	0.41					
LMWN (%)		1.7×10^{-5}	0.00058	0.0052	0.22	0.0095

water depth categories, with higher CDOC (%) in surface waters and higher HOC (%) in local and regional GW.

A comparison of sample depth categories at the non-sedimentary peat and peat sites are shown separately in Fig. 4B and Fig. 4C, respectively. At the non-sedimentary peat sites there is a transition in DOC characteristics on RDA1 from surface and hyporheic zone water which contain higher BP (%), CDOC (%), HS (%), HS aromaticity and molecular weight, and DOC concentrations to local and regional GW which contain higher LMWN (%), BB (%) and HOC (%). At sites that contain peat layers, the change in DOC character with sample depth is less clear, with regional GW and surface waters containing higher LMWN (%) and BP (%) than local groundwater which appears to contain higher HS (%), HS molecular weight and aromaticity. We propose the reason for this difference is less processed organic matter inputs, likely from the peat layers are impacting local GW DOC at these sites. Overall this suggests that overall groundwater DOC composition is less controlled by processing with depth at the peat sites, a conclusion which is supported by a higher% variation explained by the first and second constrained components for non-peat sites (19.6% and 6.5% respectively) compared to the peat sites (11.7% and 6.2% respectively) as shown in Figure S2 in the supplementary material.

4. Discussion

We have presented what is, to our knowledge, the largest dataset of groundwater DOC character using LC-OCD. Our samples come from multiple sites and over several years of sampling. Seasonal variation such as changes in evaporation, rainfall and soil moisture could lead to changes in groundwater DOC concentration and composition, particularly in surface waters and hyporheic samples which are more influenced by this shorter-term variability. Groundwater samples will tend to display less seasonal variability in DOC concentration and character compared to surface waters and hyporheic samples due to the relative stability over time of the environmental conditions that drive changes in DOC (groundwater DOC experiences constant darkness, little change in temperature, more evolved and stable water chemistry). Further discussion of seasonal variability in samples from the same sites is available in the supplementary material of McDonough et al. (2020b). In this study at Anna Bay we observe a significantly lower median value for DOC, HOC, hydrophilic, HS, BB and LMWN concentrations in the hyporheic samples in the autumn samples compared to the summer samples using one-tailed Wilcoxon rank sum tests (all $p > 0.005$). However, due to the sampling locations not being

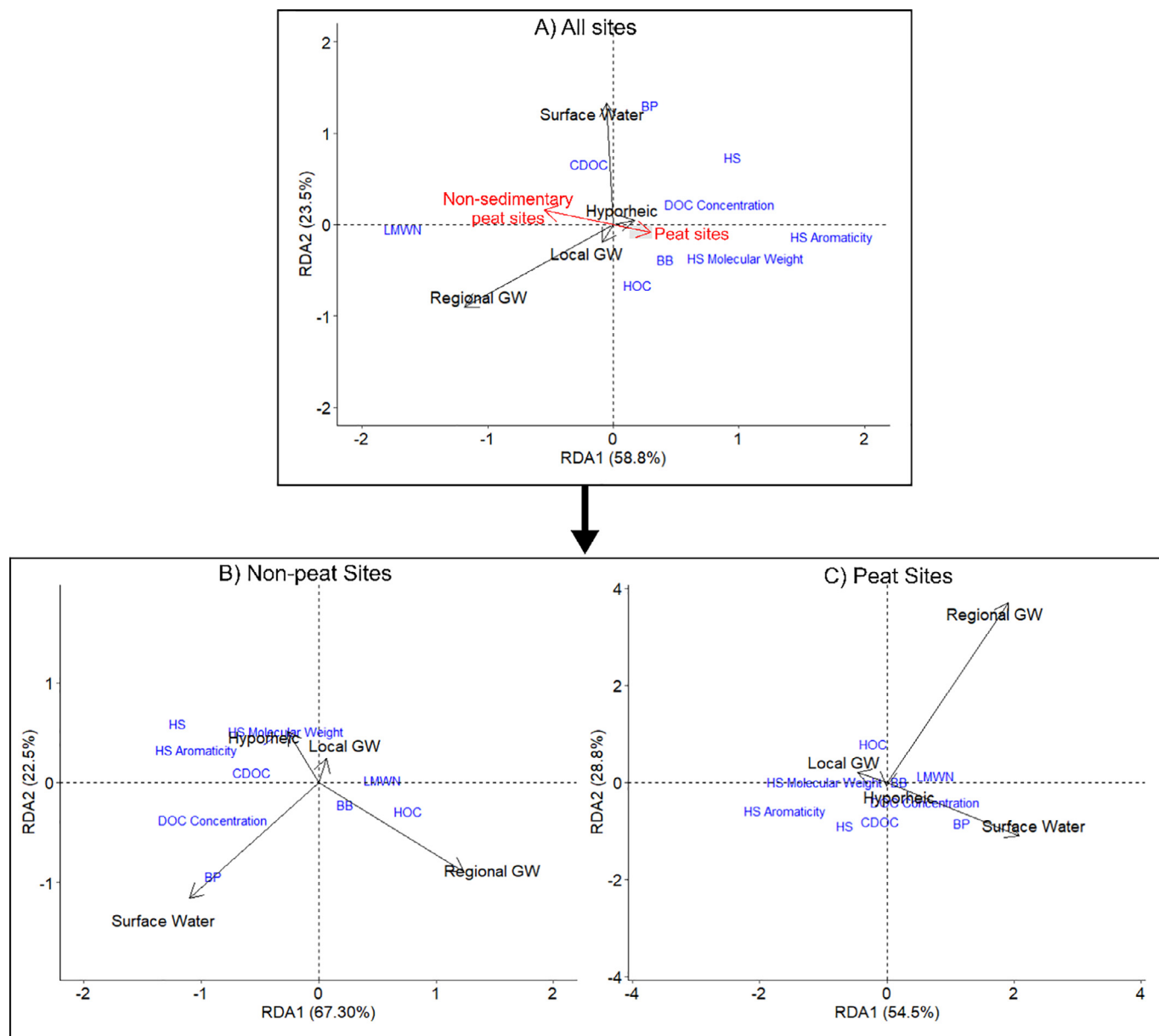


Fig. 4. Redundancy analysis using non-peat/peat sites and sample depth category (surface, hyporheic, local groundwater (GW) and regional GW) as variables explaining LC–OCD fraction concentrations for A) all sites, B) non-sedimentary peat sites (Maules Creek, Rottneest and Wellington), and C) peat sites (Anna Bay and Thirlmere Lakes). Only samples containing aromaticity and molecular weight data were included in the analyses ($n = 216$). DOC fractions shown in the RDA are percent concentrations, DOC concentration is in ppb, HS molecular weight and HS aromaticity are in grams/mol and $L/(mg \cdot m)$ respectively and are calculated as per Huber et al. (2011).

consistent between the different trips (for example, the summer dataset at Anna Bay contained more local groundwater samples while the autumn dataset contained more hyporheic samples) it is difficult to observe any seasonal trends in the data. We note however that the data represents a range of seasons and sampling dates at each site and thus this seasonal variability would be captured in the analyses presented for the full dataset, and the sedimentary peat layers and non-sedimentary peat subgroups. Overall, the differences between the sites and with depth are larger than these seasonal differences.

In all environments (with the exception of Rottneest Island) there is a decline in the higher molecular weight fractions as water flows through the subsurface (i.e. hyporheic to local and regional groundwater), such as BP and HS, which is suggestive of organic matter processing. HS have been shown to preferentially sorb compared to the other LC–OCD fractions on various aquifer materi-

als (Oudone et al., 2019). The decrease in HS in both concentration and relative abundance therefore indicate sorption of organic matter as water flows through the subsurface at all sites. This is also supported by the work by (Chapelle et al., 2016) who proposed sorption as one of the mechanisms for the removal of dissolved organic matter along flowpaths in aquifers. BP are predominately present in the surface water samples and not in the groundwater samples. We hypothesise this is due to the organic matter that is produced through microbial activity by a range of algae and bacteria present in surface water (Khan et al., 2019; Li et al., 2015; Villacorte et al., 2015; Zheng et al., 2009).

At Rottneest Island some of the samples are influenced by salt-water intrusion (see supplementary material for details). At this site, we see an increase in DOC and HS concentrations along the flowpath of fresh (local GW) to old seawater samples (Table 2). In comparison to the regional groundwater samples at the other

sites, the old seawater samples at Rottnest Island have lower relative abundances of HOC and LMWN, with a higher relative abundance of HS (Fig. 2).

The composition of the local groundwater samples that have interacted with sedimentary peat layers (Thirlmere Lakes and Anna Bay) are similar with high relative abundance and concentration of HOC compared to the other sites, however, there are differences in the abundance of the HS and LMWN fractions. The hyporheic samples at both sites have similar relative abundances which suggests that these differences are not due to any surface processes such as photodegradation. Instead we hypothesise that there is an additional source in the groundwater at Thirlmere Lakes, which has not interacted with the peat layers and has undergone more DOM processing resulting in lower HS and higher LMWN abundances (McDonough et al., 2020b).

Hydrophilic / HOC ratios are important in determining the treatability of water, with lower values suggesting easier treatability due to higher concentrations of non-polar HOC. Median hydrophilic / HOC ratios for United Kingdom and United States surface waters has been estimated at approximately 1.22 (Ghernaout 2014). Our LC-OCD results show highly variable hydrophilic / HOC ratios in groundwater at all sites, with 9.6% and 25.6% of samples at sedimentary peat layers and non-sedimentary peat sites respectively containing only hydrophilic DOC. We identify average hydrophilic / HOC ratios of 4.35 ± 3.76 and 7.53 ± 5.32 at peat and non-sedimentary peat sites respectively where both hydrophilic DOC and HOC are present ($p = 0.0021$). The results show that the presence of sedimentary organic matter is the main control on groundwater DOC composition (as demonstrated by RDA1 on Fig. 4a), with a secondary control by sample depth at the non-sedimentary peat sites. DOM at sites containing sedimentary peat layers has a higher abundance of HOC, HS of higher molecular weight and overall DOC concentrations being between 2 and 15 times higher than fractured rock and alluvial environments for the equivalent depth. Pre-treatment by coagulation/flocculation, sedimentation or sand filtration which have high removal efficiency for HOC (Ghernaout 2014; Krzeminski et al., 2019; Sharp et al., 2006) may be more suitable treatment methods for these DOM characteristics. The presence of aged organics at depth can also lead to a higher abundance of HOC, such as at Maules Creek, however the overall HOC concentration is significantly lower than at sites containing sedimentary peat layers, and hence is unlikely to require pre-treatment. In fractured rock and alluvial groundwater environments, DOC is lower in concentration and has a higher proportion of LMWN, and HS of lower molecular weight than HS in environments containing peat layers. DOC in these groundwaters may therefore be more efficiently removed by ozonation, biologically activated carbon (BAC) filtration, PAC, SIX or magnetic ion exchange resin which can remove lower molecular weight and hydrophilic fractions (Andersson et al., 2020; Krzeminski et al., 2019; Mergen et al., 2009; Shutova et al., 2020).

5. Conclusions

This study represents the largest dataset of chromatographic DOM in surface and groundwater samples ($n = 239$) using liquid chromatography organic carbon detection (LC-OCD). Our data represents four depth categories (surface water, hyporheic zone water, local groundwater, and regional groundwater) from five different sites in Australia. Our findings suggest that DOM composition can be highly variable and highlight the complexity and variability of DOM inputs and processing mechanisms of various size fractions in different environments. While we observe higher percentages of difficult-to-treat hydrophilic organic carbon in groundwater from non-sedimentary peat environments, the DOC concentrations tend to be lower. Our findings indicate that the presence

of sedimentary organic matter strongly influence the character of groundwater DOM, resulting in different requirements for removal of DOC from these environments. Depth also affects groundwater DOC concentration and composition with sedimentary organic carbon inputs in peatland environments resulting in inputs of higher molecular weight, higher aromaticity DOM in shallow groundwater. In all environments DOC concentration and high molecular weight fractions such as BPs decline with groundwater depth due to processing through the subsurface.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at [doi:10.1016/j.watres.2020.116422](https://doi.org/10.1016/j.watres.2020.116422).

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