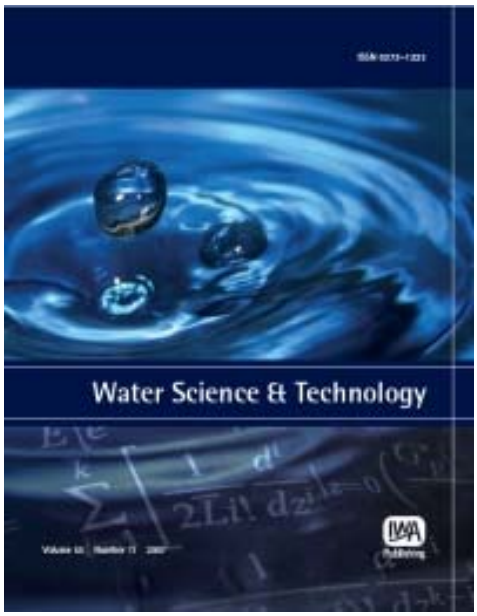

Distinguishing stage 1 and 2 reverse osmosis permeates using fluorescence spectroscopy

S. Singh, R. K. Henderson, A. Baker, R. M. Stuetz and S. J. Khan

ABSTRACT

Fluorescence excitation-emission matrix (EEM) spectroscopy was used to distinguish between two stages of reverse osmosis (RO) permeates as the first step towards investigating the potential application of fluorescence as a monitoring tool for membrane performance. The signal response of several fluorescence peaks present in Stage 1 and Stage 2 RO permeates of an advanced water treatment plant were compared. The humic-like fluorescence region was found to have the largest percentage difference between stages and therefore was the most appropriate for enabling differentiation. Increases in humic-like fluorescence did not correlate with increases in conductivity or dissolved organic carbon measurements. This suggests that fluorescence is a more selective and sensitive method for monitoring the organic composition of RO permeates than established methods. Fluorescence is therefore a promising tool for improved water quality monitoring of RO permeates.

Key words | conductivity, fluorescence, reverse osmosis, water quality monitoring

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INTRODUCTION

The use of reverse osmosis (RO) systems in water treatment has gained popularity over recent years for applications including desalination and recycling of treated wastewater effluent. Monitoring membrane performance is critical to ensure water of adequate quality is produced. Total organic carbon (TOC) and conductivity are established methods for monitoring water quality and assessing membrane integrity (Adham *et al.* 1998; Kumar *et al.* 2007). However, conductivity has been found to be less sensitive than TOC (Adham *et al.* 1998) and not a good predictor of viral rejection (Kitis *et al.* 2003). Overall, these studies indicate membrane performance is better monitored by targeting the organic component, as opposed to small ions via conductivity.

Analysis of fluorescent dissolved organic matter (DOM) has been identified as a potential monitoring tool for membrane performance (Henderson *et al.* 2009). A significant proportion of DOM in freshwater fluoresces and can provide a good indication of water quality. Coble (1996) categorized freshwater fluorescent peaks as humic-like (A: λex/em = 237–260/400–500 nm, C: λex/em = 300–370/400–500 nm), tyrosine-like (B: λex/em = 225–237/309–321 nm and λex/em 275/310 nm) and tryptophan-like (T_1:λex/em = 275/340 nm, T_2: λex/em = 225–237/340–381 nm). Subsequent studies reclassified peak C into two smaller peaks; C_1 (λex/em = 320–340/410–430 nm) and at C_2 (λex/em = 370–390/460–480 nm) (Baker 2001; Baker *et al.* 2008). Fluorescence has been utilized to monitor water quality and pollution in rivers (Hudson *et al.* 2007), control processes in sewage treatment works (Reynolds & Ahmad 1997; Ahmad & Reynolds 1999; Hudson *et al.* 2007), the formation of disinfection byproducts in drinking water (Hua *et al.* 2007), oil spills in water (Lambert 2003) and specific pollutants in industrial wastewater (Kuzniz *et al.* 2007). The appeal of fluorescence spectroscopy is that it is a rapid, yet sensitive method that requires no prior sample preparation. Modern fluorescence spectrophotometers allow data to be generated as 3-dimensional contour graphs to form a matrix (excitation × emission × intensity); termed

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Table 1 | Plant influent and RO feed water quality range for sampling period

Feed water source	Conductivity ($\mu S\,cm^{-1}$)	DOC ($mg\,L^{-1}$)	pH	Turbidity (NTU)
Plant influent	620–1314	8.98–11.79	6.74–7.85	0.90–2.30
RO feed	628–1318	3.06–9.96	6.84–8.06	0.10–0.31

excitation-emission matrix (EEM) (Mobed *et al.* 1996). An EEM can be generated in less than one minute depending on the sensitivity required, enabling rapid fluorescence peak identification.

Interpreting fluorescence EEMs is a complex process due to variability in the fluorophore number, structure and composition. Several data interpretation techniques have been used to analyse natural organic matter. Traditionally, simple techniques utilizing peak intensities and ratios have been used to monitor organic matter in rivers, lakes, oceans (Henderson *et al.* 2009). An alternative technique–Fluorescence Regional Integration (FRI)–devised by Chen *et al.* (2003), involves the division of wastewater EEMs into five regions based on common freshwater fluorescent peaks (Table 1). Each region is integrated and normalised to provide a fluorescence volume for that region. More complex chemometric techniques, including principal components analysis (PCA), partial least squares regression, principal filter analysis and parallel factor analysis (PARAFAC), are becoming popular in analyzing natural organic matter fluorescence (Henderson *et al.* 2009).

This study investigates the ability of fluorescence EEM spectroscopy to distinguish between two different stages of RO permeates in order to determine the potential for using fluorescence as a tool for monitoring membrane performance. Suitable fluorescence excitation-emission region for monitoring membrane performance are identified. Data interpretation is conducted using FRI and a modified version of this technique.

MATERIALS AND METHODS

Site description

Samples were collected from the Water Reclamation and Management Scheme (WRAMS) water treatment plant (WTP), Sydney Olympic Park Authority (SOPA), in Australia. The WTP has both continuous-flow microfiltration (CMF) and RO membrane filters. The plant can treat a combination of stormwater and secondary effluent with a total capacity of 7.5 ML per day. All the water is filtered through the CMF units while a maximum of 2 ML per day goes through the RO units. The WTP has two RO skids, comprising of six stage 1 modules and three stage 2 modules. The RO permeates are blended with the CMF water prior to distribution to produce recycled water of acceptable quality. Generally the skids are operated sequentially with the change over occurring every 10 hours.

Sample collection

Sample collection was conducted over a period of eight weeks from August to October, 2008. RO permeates were collected (in triplicate) in 50 mL polypropylene bottles, refrigerated and analysed within 4 days. Samples were obtained from all elements of the operational skid (Skid A: weeks 2, 4, 5, 6, 8; Skid B: weeks 1, 3, 6, 7) except in week 6 when both skids were operating. For week 6, samples were taken from three stage 1 modules and one stage 2 from each skid. Plant influent and RO feed quality for the sampling period is detailed in Table 1.

Analytical methods

Fluorescence EEMs were acquired using a bench Varian Cary Eclipse fluorescence spectrophotometer. Excitation wavelengths were monitored from 200 nm to 400 nm in 5 nm increments, while emission wavelengths were monitored from 280 nm to 500 nm in 2 nm increments. Excitation and emission slits were set at 5 nm, scan speed at 9,600 nm min^{-1} and photomultiplier tube (PMT) voltage set to 1,000 V. Three averages were recorded for each scan. Temperature was kept constant at 25 ± 0.5°C. Blank water

Table 2 | FRI EEM regions, according to Chen *et al.* (2003)

Region	Excitation (nm)	Emission (nm)	Description
I	220–250	280–332	Aromatic proteins I
II	220–250	332–380	Aromatic proteins II
III	220–250	380–500	Fulvic acid-like
IV	250–400	280–380	Microbial by-products
V	250–400	380–500	Humic acid-like

Table 3 | Modified EEM FRI regions

Region	Excitation (nm)	Emission (nm)	Description	No. of data points per region	Projected excitation-emission area (nm²)
A	235–260	400–440	Humic acid-like	126	1,000
C_1	305–340	406–430	Humic acid-like	104	840
T_2	235–250	340–382	Tryptophan-like	88	330
Summation				318	2,170

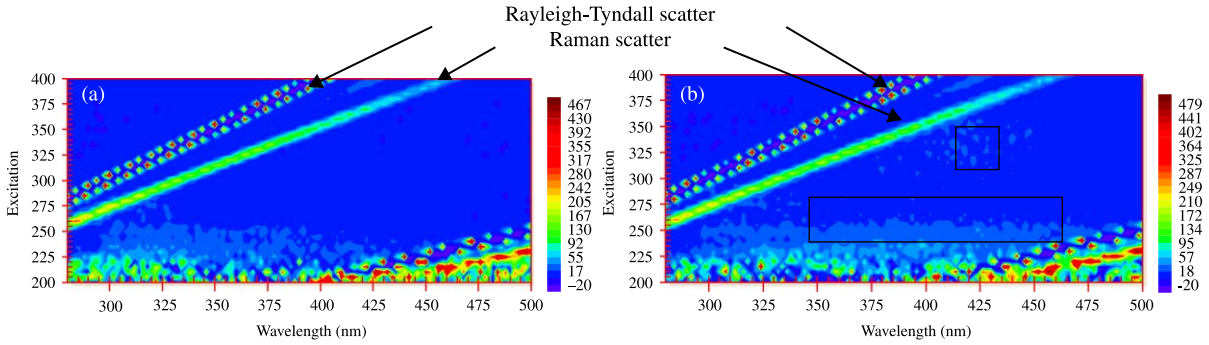

Figure 1 | Typical RO permeate EEMs for (a) stage 1 and (b) stage 2, illustrating an increase in fluorescence intensity for stage 2. Rayleigh-Tyndall and Raman scatter lines which do not relate to water quality are marked. Subscribers to the online version of Water Science and Technology can access the colour version of this figure from http://www.iwaponline.com/wst.

scans were obtained using a sealed MilliQ water cell (Varian) and subtracted from all fluorescence spectra. The intensity of all EEM spectra was normalized by using the Raman peak of water at excitation 348 nm.

UV absorption spectra (200–600 nm) were obtained using a Varian Cary 50 UV Absorption Spectrometer. Conductivity and pH levels were measured with a HACH HQ14d portable meter. Dissolved Organic Carbon (DOC) values were obtained using liquid chromatography–organic carbon and nitrogen detection (LC-OCD Model 8, DOC-Labor, Germany) equipped with a TSK HW 50S (250 mm × 20 mm) column.

Data analysis

Quantification of EEM spectra was first performed following the Fluorescence Regional Integration (FRI) technique (Chen *et al.* 2003). In brief, the EEM spectra were divided into five regions characteristic of specific components of DOM, as detailed in Table 2. The volume of fluorescence intensity under each region was calculated and normalized by multiplying the inverse of the fractional projected area under each region (MF_i). The normalized volume ($\Phi_{i,n}$) was then converted to percent fluorescence response ($P_{i,n}$) using Equations (1) and (2). Converting data to percent fluorescence response ($P_{i,n}$) had two major advantages. First, the relative proportions of the regions within the EEM could be compared. Secondly, the changes in relative proportions of the regions in the two RO stages could be compared.

$$\phi_{T,n} = \sum_{i=1}^{5} \phi_{i,n} \quad (1)$$

Table 4 | Percent fluorescence response using FRI for stage 1 and 2 RO permeates

Region	Percent fluorescence response averaged over 8 weeks			
	Stage 1		Stage 2	
	Percentage	Std dev	Percentage	Std dev
I	34.4	7.4	26.6	4.8
II	30.8	11.0	30.1	7.6
III	17.9	6.5	23.4	5.2
IV	12.5	4.9	11.5	4.8
V	4.4	1.3	8.3	1.1
Total	100		100	

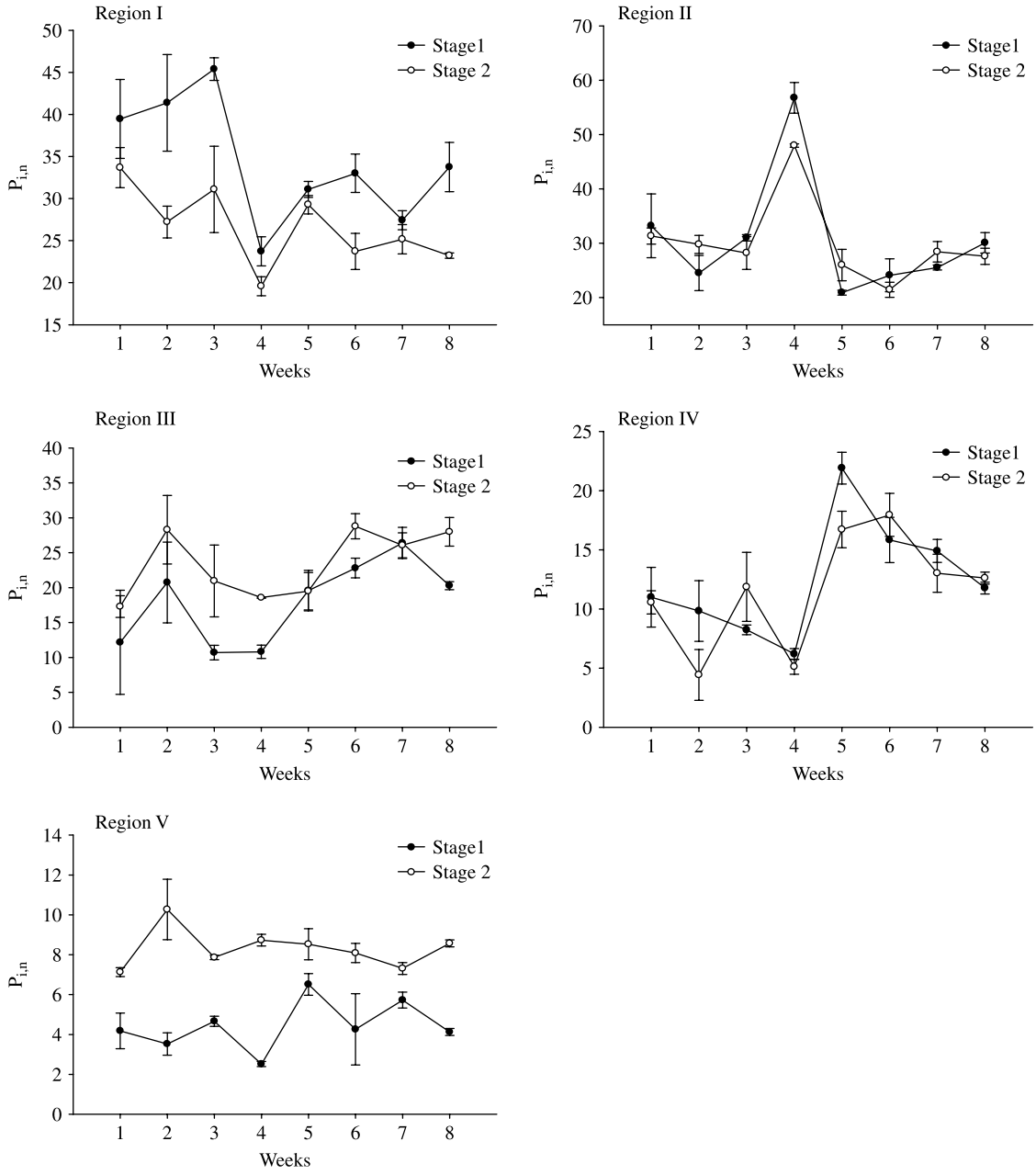

Figure 2 | Fluorescence percent response using the FRI method for regions I–V for stage 1 and 2 RO elements over the 8 week sampling period.

$$P_{i,n} = \phi_{i,n}/\phi_{T,n} \times 100 \quad (2)$$

The FRI method was then modified to obtain a second quantification by altering the region boundaries. The modified FRI method selectively reapplied the FRI technique to smaller areas to further isolate the regions of interest. Specifically, Peaks A, C_1 and T_2 were encompassed based on literature reports and observed fluorescence. The modified FRI did not utilize the entire EEM; volume integration was conducted according to Equation (3) within specific peak areas (Table 3).

$$\phi_{T,n} = \phi_A + \phi_{C1} + \phi_{T2} \quad (3)$$

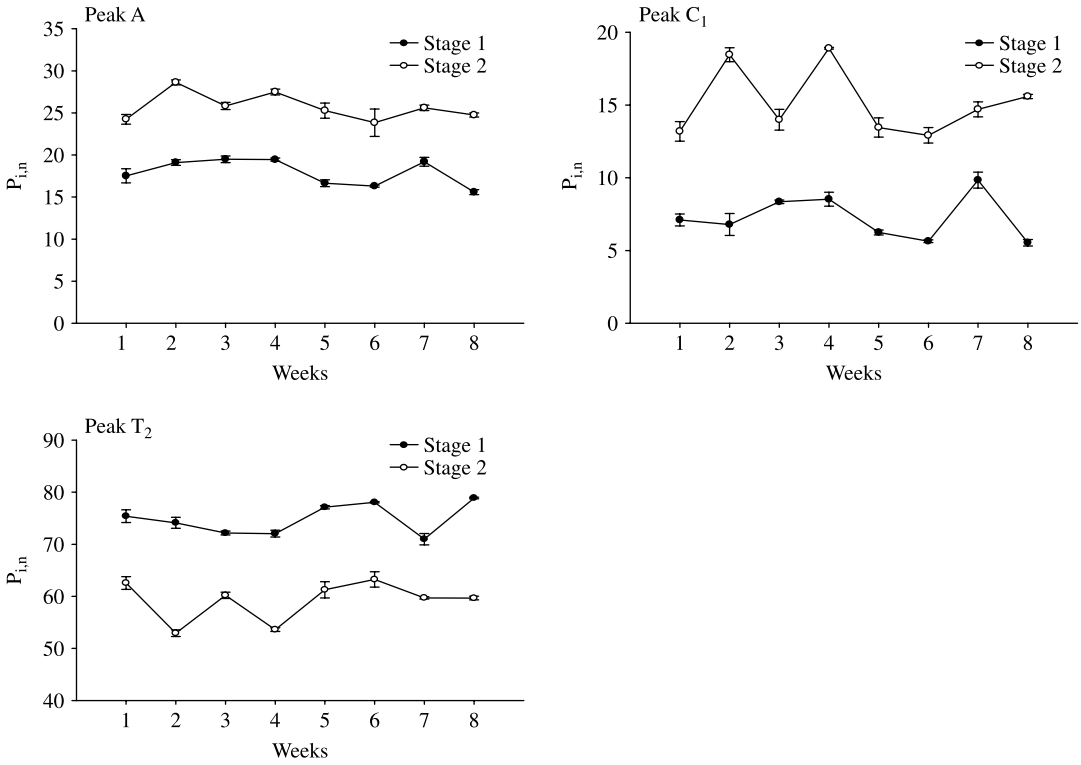

Figure 3 | Fluorescence percent response using the modified-FRI method for Peak A, Peak C_1 and Peak T_2 for stage 1 and 2 RO elements over the 8 week sampling period.

RESULTS AND DISCUSSION

Typical fluorescence EEMs are shown in Figure 1, which illustrates that fluorescence intensities increased for stage 2 relative to stage 1. Results were first interpreted using the FRI technique. A comparison of the five regions averaged over 8 weeks showed that while fluorescence composition was variable, the bulk of the fluorescence was concentrated in regions I–III (Table 4). Regions I and II (aromatic proteins) constituted approximately 30% of the total fluorescence. Region III (fulvic acid-like) and Region IV (microbial by-products) comprised 18% and 12.5% respectively. Region V had the lowest composition of fluorescence (4% for stage 1 and 8% for stage 2 permeates). On comparison of the fluorescence percent response of the two RO stages (Figure 2), it was observed that only monitoring region V enabled consistent differentiation (Figure 2).

The regions of interest were then narrowed according to the modified-FRI method. On comparison of fluorescence volumes prior to conversion to percent fluorescence response (data not shown), it was observed that Peaks A and C_1 consistently demonstrated no overlap between stage 1 and stage 2 permeates during the sampling period while T_2 exhibited partial overlap. The fluorescence volume for all the regions increased in stage 2 but Peaks A and C_1 exhibited a larger increase in fluorescence compared to T_2. This became evident when the data was converted to percent fluorescence response (Figure 3), where percent fluorescence response for stage 1 RO permeates was greater

Table 5 | Percent fluorescence response using modified FRI for stage 1 and 2 RO permeates

Peak	Percent fluorescence response averaged over 8 weeks			
	Stage 1		Stage 2	
	Percentage	Std dev	Percentage	Std dev
A	17.9	1.6	25.7	1.7
C1	7.3	1.5	15.1	2.3
T2	74.8	3.0	59.2	3.8
Total	100		100	

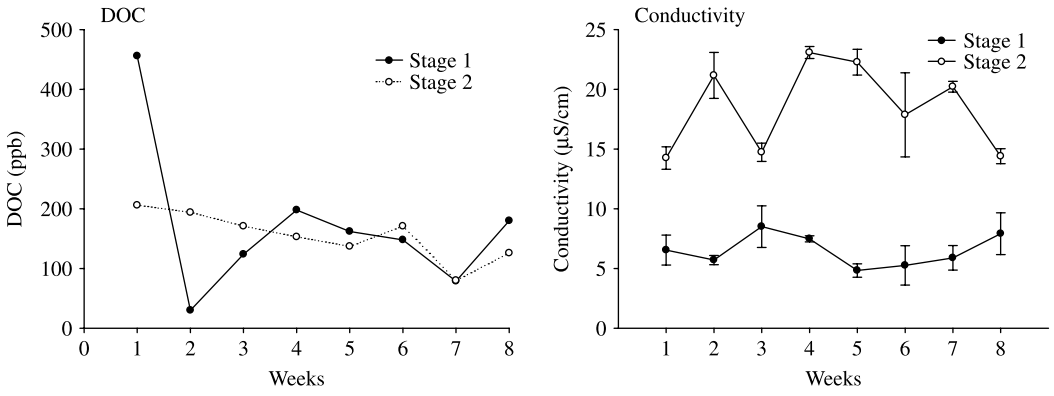

Figure 4 | Conductivity and DOC results for Stage 1 and 2 over the 8 week sampling period.

than stage 2 for peak T_2. On average, the percent fluorescence response of component Peaks A and C_1 increased from approximately 17.9% and 7.3% to 25.8% and 15.1% respectively, while Peak T_2 decreased from 74.8% to 59.2% (Table 5). The modified-FRI fluorescence results indicate that humic-like organics (peak C_1) are the most representative of the changes in permeate water quality and are most appropriate for monitoring purposes.

DOC results ranged from less than $50\,\mu g\,L^{-1}$ to $470\,\mu g\,L^{-1}$ and it was not possible to distinguish between stage 1 and 2 permeates (Figure 4). In contrast, conductivity results were consistently greater for stage 2 relative to stage 1 (Figure 4). Interestingly, conductivity and DOC results did not follow similar trends. For example, for stage 2, DOC values were three times higher in week 1 than in week 4 while conductivity was 3.5 times lower, thus indicating that organics are penetrating, or leaching from, the membrane, while salt removal is good. Similarly, while Peak C_1 and conductivity exhibited relatively similar trends for the first six weeks (Figures 3 and 4), in week 7, conductivity for stage 1 permeates showed a decrease while fluorescence response of Peak C_1 was increased. This trend was reversed in week 8. These examples highlight that conductivity alone cannot give an overall indication of process performance.

Conductivity always demonstrated a significantly higher percent differentiation between stage 1 and 2 in comparison to fluorescence results (Figure 5); however, measuring conductivity did not give an indication of the relative DOM permeation. The modified-FRI method was more sensitive to changes in permeate water quality than the FRI method. For example, Peaks A and C_1 had a percent difference of approximately 4% greater than region V (Figure 5). One reason for the improvement in differentiation is that the FRI method incorporates Raman and Rayleigh scatter. While blank subtraction significantly reduces the fluorescence attributed to scatter lines some residual fluorescence remains, contributing to the overall volume. The modified FRI regions do not incorporate these scatter lines and thus fluorescence volumes are unaffected.

The humic-like fluorescence did not correlate with DOC trends (Figure 3 and 4). Both are measures of organic matter content but while DOC relates the total organic matter present, fluorescence results only represent fluorescent organic concentration in the RO permeates. The fluorescence results show that by focusing on a specific fluorescent component–humic-like fluorescence–distinguishing between stage 1 and 2 RO permeates can be achieved. Interestingly, an increase in humic-like fluorescence, indicating a change to poorer water quality, was not always reflected by a similar trend in conductivity

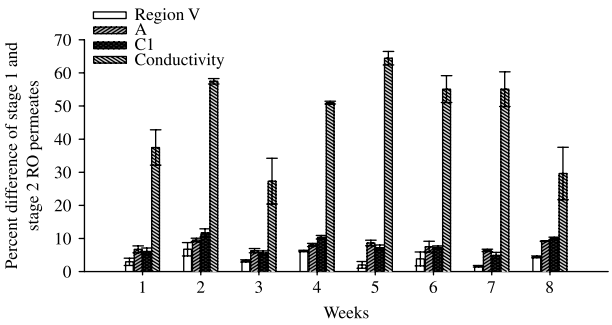

Figure 5 | Percent signal difference between Stage 1 and 2 RO permeates for fluorescence and conductivity.

(Figures 3 and 4). This observation is related to the RO membrane rejection mechanisms. Solute rejection in RO membranes is influenced by solute properties, membrane properties and feed water composition (Bellona *et al.* 2004). Low molecular weight neutrals are known to permeate through RO membranes while acidic organics tend to be rejected due to electrostatic repulsion (Bellona *et al.* 2004). The observation of increased humic-like fluorescence in the RO permeate of Stage 2 suggests that electrostatic repulsive forces are overcome by increased osmotic pressure.

CONCLUSIONS

Monitoring humic-like fluorescence consistently differentiated between stage 1 and 2 RO permeates using both FRI and modified-FRI methods, where the latter gave the best separation. Increases in humic-like fluorescence were not always correlated with DOC or conductivity results, indicating that fluorescence was a more sensitive method for the detection of changes in the organic composition of the permeate. Further investigation is now required to develop an on-line method that can be applied in the field to monitor RO membrane underperformance/failure.

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