Effect of increasing bromide concentration on toxicity in treated drinking water

Emma Sawade, Rolando Fabris, Andrew Humpage and Mary Drikas

ABSTRACT

Research is increasingly indicating the potential chronic health effects of brominated disinfection by-products (DBPs). This is likely to increase with elevated bromide concentrations resulting from the impacts of climate change, projected to include extended periods of drought and the sudden onset of water quality changes. This will demand more rigorous monitoring throughout distribution systems and improved water quality management at water treatment plants (WTPs). In this work the impact of increased bromide concentration on formation of DBPs following conventional treatment and chlorination was assessed for two water sources. Bioanalytical tests were utilised to determine cytotoxicity of the water post disinfection. Coagulation was shown to significantly reduce the cytotoxicity of the water, indicating that removal of natural organic matter DBP precursors continues to be an important factor in drinking water treatment. Most toxic species appear to form within the first half hour following disinfectant addition. Increasing bromide concentration across the two waters was shown to increase the formation of trihalomethanes and shifted the haloacetic acid species distribution from chlorinated to those with greater bromine substitution. This correlated with increasing cytotoxicity. This work demonstrates the challenges faced by WTPs and the possible effects increasing levels of bromide in source waters could have on public health.

Key words | chlorination, coagulation, cytotoxicity, disinfection by-products, drinking water

Emma Sawade (corresponding author)
Rolando Fabris
Andrew Humpage
Mary Drikas
Australian Water Quality Centre,
South Australian Water Corporation,
250 Victoria Square,
Adelaide,
SA 5000,
Australia
E-mail: *emma.sawade@sawater.com.au*

INTRODUCTION

When chemical disinfection is applied to drinking water, compounds form that may pose a risk to the health of humans and aquatic organisms. Chlorine is commonly used as the primary disinfectant in drinking water supplies across Australia, and in many regions throughout the world. While chlorine's disinfection ability has provided substantial public health benefits, its interaction with natural organic matter (NOM) and inorganic precursors (e.g. iodide and bromide) that may be present can generate hundreds of disinfection by-products (DBPs) (Agus & Sedlak 2010; Wang *et al.* 2010). Although chlorinated water has been linked with adverse health effects and over 600 DBPs have been identified to date (Malliarou *et al.* 2005), the toxicity of only approximately 20 of them has been investigated in some detail (Sadiq & Rodriguez 2004; Al-Mudhaf *et al.* 2010). Consistent efforts have been made to determine identities and toxicities of various DBPs, especially those of trihalomethanes (THMs) and haloacetic acids (HAAs), and to model their formation and control their occurrence (Roccaro *et al.* 2008).

In general, higher THM and HAA concentrations are produced with increasing disinfectant dose, pH, temperature and the precursor concentration (Twort & Brandt 2000; Uber 2003; Sadiq & Rodriguez 2004; El-Hassan & Al-Sulami 2005). The exception for HAA formation is that this is favoured at lower pH values (Rodriguez & Serodes 2005). Many researchers have observed that bromide incorporation into DBPs is favoured when the molar concentration of chloride and bromide is similar (Wang *et al.* 2010). The bromide ion (Br^-) is an important DBP precursor as its concentration affects the kinetics and nature of the distribution of adsorbable organic halides (AOX) and DBPs formed (El-Hassan & Al-Sulami

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2005; Obolensky & Singer 2008; Agus *et al.* 2009). The reaction of hypochlorous acid (HOCl) with bromide produces hypobromous acid (HOBr), which reacts faster with NOM than HOCl, incorporating bromine as well as chlorine into precursor organic compounds (Nikolaou & Lekkas 2001; Westerhoff *et al.* 2004; Hua *et al.* 2006; Wang *et al.* 2010). Hence, in the presence of bromide, elevated production of brominated and mixed chloro-bromo-substituted DBPs occurs. Reported factors for the reaction rates of halogen incorporation into organic matter have ranged between three times (Heeb *et al.* 2014) and ten times (Westerhoff *et al.* 2004) faster bromide reactivity than chlorine. Studies using ultraviolet-visible spectroscopy and solid-state Carbon-13 Nuclear Magnetic Resonance spectroscopy to investigate these reaction mechanisms found that aqueous bromine substitutes into organic structures, whereas chlorine tends to cleave carbon bonds, thus having a more significant impact on NOM structure (Westerhoff *et al.* 2004). Bromo-derivatives are formed in the presence of aliphatic precursors whereas chloro-substitution occurs preferentially in the case of aromatic compounds. Therefore, the speciation and concentration of DBPs formed during chlorination are mainly dominated by the ratio of bromide to reactive NOM as well as the ratio of bromide to chlorine concentrations (Westerhoff *et al.* 2004).

The occurrence of bromide in surface and ground water typically reflects intrusion from marine sources (bromide coexists with chloride in salt), or is derived from salts created by human activity (e.g. halide applied to roads as anti-freeze during winter) (Weinberg *et al.* 2003). The highest concentrations of bromide exist in arid areas with high evaporation rates such as south-western Australia, western United States and the Mediterranean (Agus *et al.* 2009). A survey of water supplies showed that in arid regions, concentrations of bromide in freshwater sources, such as in the Helena River in southwest Australia ($Br^- = 700\ \mu g/L$), can be more than 20 times higher than the median concentration ($Br^- = 35\ \mu g/L$) measured for a wide range of surface water bodies worldwide (Agus *et al.* 2009).

It is worth noting that while optimised water treatment processes (i.e. coagulation and adsorption) remove organic compounds, they do not similarly remove the bromide ion, hence the bromide to dissolved organic carbon (DOC) ratio increases after those treatments, which consequently increases the fraction of brominated species of THMs and HAAs formed (Roccaro & Vagliasindi 2009). Thus, even if the total amount of DBP is reduced, the overall toxicity may not be. Bioanalytical methods targeting key biological effects are well suited to assess this risk. Unlike most chemical analyses that incorporate a fractionation stage prior to the detection stage, bioassays measure the biological effect of the complete chemical mixture in a sample. They are therefore particularly useful for screening applications or, as in this case, when all active compounds have not been identified and so cannot be targeted by chemical analysis (Leusch *et al.* 2014). Brominated species of THMs and HAAs have been shown to exhibit stronger cytotoxic, genotoxic and mutagenic effects in cell-based assays than their chloride-containing analogues (DeMarini *et al.* 1997; Pegram *et al.* 1997; Plewa *et al.* 2002; Ross & Pegram 2003). Similarly, brominated DBPs are generally more genotoxic and carcinogenic than their chlorinated counterparts when tested in animal studies (Richardson *et al.* 2007). Thus, cell-based bioassays have been used both to determine the relative potency of individual DBPs in order to prioritise them for further research (Muellner *et al.* 2007; Plewa *et al.* 2008; Laingam *et al.* 2012) as well as to assess treatment processes for production of harmful DBPs (Claxton *et al.* 2008; Neale *et al.* 2012).

The aim of this work was to evaluate the impact that increased bromide concentration would have on the formation of THMs, HAAs and total DBPs formed, as measured by AOX, following conventional treatment and chlorination. In addition, the impact on the cytotoxicity of the water after disinfection was assessed by using bioanalytical tests.

MATERIALS AND METHODS

Water sources

Source water (150 L) from two locations, a reservoir in Western Australia (WA) and the inlet of a regional water treatment plant (WTP) located on the River Murray in South Australia (SA), were collected and treated in the laboratory. The water sources were selected based on their differing bromide and DOC content; the Western Australian water had high bromide (0.60 mg/L) and high DOC concentration (13.8 mg/L) while the South Australian water had

low bromide (0.07 mg/L) and moderate DOC concentration (7.2 mg/L).

Treatment and disinfection conditions

Coagulation/filtration jar testing was undertaken using a PB-900 6-paddle gang stirrer (Phipps & Bird, USA). Aluminium sulphate (alum) doses of 25, 50, 75, 100, 125, 150 mg/L as $Al_2(SO_4)_3.18H_2O$ was applied for the Western Australian water and 30, 40, 60, 70, 80, 100 mg/L for the South Australian water source. From this, the coagulant dose (optimum) that produced water that met aesthetic water quality parameters, $\leq$0.1 nephelometric turbidity units (NTU) turbidity and <10 hazen units (HU) colour, was determined. The jar test conditions were 1 minute at 200 rpm flash mix, 14 minutes at 20 rpm slow mix and 15 minutes settling in 2 L square form Gator Jars. Water was gravity filtered through 11 μm nominal pore size paper filters (No. 2, Advantec, Japan). For simplicity no pH adjustment was carried out.

Water treated using the optimum coagulation conditions, 125 mg/L for WA and 70 mg/L for SA, was spiked with additional bromide to achieve higher bromide concentrations. Samples were then disinfected using 72-hour simulated distribution system (SDS) tests at 25 °C, representing summer conditions. Chlorine was dosed at a concentration to replicate the 72-hour chlorine demand +0.5 mg/L (as residual), representing a realistic chlorine dose at the WTP. Samples were taken at regular intervals (0.5, 4, and 72 hours) for chlorine, THMs, HAAs and AOX. All samples were evaluated using the WIL2-NS cytotoxicity bioassay.

Analytical tests

Samples for DOC, true colour and UV_{254} absorbance analyses were filtered through 0.45 μm pre-rinsed membranes. UV_{254} absorbance was measured through a 1 cm quartz cell and true colour at 456 nm through a 5 cm cell using an Evolution 60 Spectrophotometer (Thermo Scientific, USA). Colour was expressed in HU by calibration against a 50 HU Pt/Co standard according to the method of Bennet & Drikas (1993). DOC was measured using a Sievers 900 Total Organic Carbon Analyser (GE Analytical Instruments, USA). Specific UV absorbance (SUVA) was calculated as UV absorbance (per metre) divided by DOC. Turbidity measurements were conducted on a 2100AN Laboratory Turbidimeter (Hach, USA) with results expressed in NTU. Measurement of pH was performed using a portable pH meter with a sealed, gel filled reference electrode with temperature compensation (pH320, WTW, Germany). All chlorine residual and chlorine demand titrations were carried out as per Standard Method 4500-Cl (F) (APHA AWWA & WEF 1998). Aqueous solutions of bromide were prepared using 1000 mg/L stock solution of bromide and stoichiometrically calculating for the required concentration for a particular bromide dose. Volatile and semi-volatile THMs (including chloroform, dichlorobromomethane, dibromochloromethane and bromoform) were analysed using the Purge and Trap Gas Chromatographic method according to Standard Method 6232 (C) (APHA AWWA & WEF 1998), while nine HAA species containing chlorine and bromine were analysed using Micro Liquid-Liquid Extraction Gas Chromatographic – Standard Method 6521 (B). The AOX samples were analysed via the method of DIN (1985) and bromide concentrations were determined by ion chromatography according to Standard Method 4110 (B) (APHA AWWA & WEF 1998).

Cytotoxicity of water samples

Mammalian cell cytotoxicity testing was carried out using a human white blood cell (WIL2-NS) based bioassay. Suspension cultures (2.5×10^4 cells/well, 96 well plates) were treated for 24 hours and the biological endpoint measured was reduced cell viability compared to the vehicle control (1% methanol), which was expressed as per cent cytotoxicity (100 – per cent viability). Cell viability was quantified using the resazurin fluorimetric assay (Nociari *et al.* 1998; Leusch *et al.* 2014). Methylmethane sulphate (10, 20, 30, 40, 60 and 100 μg/mL) was used as the positive control.

Chlorinated samples were quenched stoichiometrically with sodium thiosulphate (based on residual Cl_2 titration) before toxicity tests were undertaken to minimise any potential impacts from both free chlorine and the quenching agent (Froscio *et al.* 2009).

Solid phase extraction using Oasis® hydrophilic-lipophilic balance (HLB) cartridges (Waters, Sydney, Australia) modified from Chapman *et al.* (2011) was used to concentrate all the treated water samples prior to toxicity testing. WA water cytotoxicity data was obtained from a 10× concentration and

SA water from a 40× concentration. These sample concentrations were based on testing of positive controls to achieve approximately 80% cytotoxicity. The chlorinated untreated waters were used as positive controls and the unchlorinated treated waters were used as negative controls. Volatile DBPs including THMs are eliminated from this assay due to sample pre-concentration (Neale *et al.* 2012).

RESULTS AND DISCUSSION

Water quality

Optimum treated water quality is shown in Table 1. The waters differed in both DOC and bromide concentrations and hence had different bromide to DOC ratios (Br:DOC).

The treated water was dosed with additional bromide to explore the effects on both the DBP formation characteristics (rate and speciation) and the effect this would have on cytotoxicity. Ratios for the SA water source were chosen based on those observed in the inlet water to the WTP over the past 5 years which have varied from 1:8 to 1:294. The treated water had a ratio of 1:52, with a bromide concentration of 0.06 mg/L and DOC of 3.1 mg/L. Additional bromide was added to achieve ratios with DOC of 1:15 and 1:8 in the treated water. The ratio of 1:8 was the highest seen in the inlet water and the ratio of 1:15 meant that the amount of bromide (0.21 mg/L) present was similar to the average observed in the inlet water over the past 5 years.

For the WA water source, ratios were chosen increasing from the current bromide concentration (1:13) up to a high bromide concentration to give us a range of ratios for the water source (1:4, bromide 1.8 mg/L). The naturally higher bromide levels of the WA water allowed for further assessment of the impact of increasing bromide, for example due to climate change. For comparison, although actual added concentrations were considerably higher in the WA water source, the ratio of 1:8 was common for both water sources.

Bromide consumption during chlorination SDS of the optimally treated water was determined by measuring the residual bromide after 72 hours chlorine contact; results are shown in Table 2. The results for the WA water source showed that not all bromide was consumed for any of the three ratios, with measurable Br residuals detectable after 72 hours chlorine contact; amount consumed 85%, 89% and 80% for Br:DOC ratios 1:13, 1:8 and 1:4, respectively. Similarly bromide residuals were detected for the ratios, 1:8 and 1:15, for the SA water source with 74% and 76% consumed, respectively, while bromide was undetectable at a Br:DOC ratio of 1:52. As HOBr is the faster reacting oxidant, complete consumption of the bromide was expected. However Wang & Huang (2006) have shown that incomplete oxidation of bromide to HOBr can occur, so all of the bromide may not have been available for reaction with DOC

Table 1 | Raw and treated water quality at optimum coagulant dose

	Reservoir WA		River SA		
Parameter	Raw	125 mg/L alum	Raw	70 mg/L alum	Units
pH	5.3	6.3	8.1	6.6	
Colour	158	5	39	3	HU
Turbidity	4.2	0.1	55	0.1	NTU
UV_{254}	0.721	0.118	0.254	0.053	abs/cm
DOC	13.8	7.2	7.2	3.1	mg/L
SUVA	5.22	1.64	3.5	1.66	abs/m/mg/L
Bromide	0.60	0.55	0.07	0.06	mg/L
Br:DOC	1:23	1:13	1:103	1:52	ratio

Table 2 | Bromide comparison across treated waters

Water source	Br:DOC ratio	Br (mg/L)	DOC (mg/L)	Cl_2 dose applied for 72 hours SDS (mg/L)	Br consumed (mg/L)	Actual Br (mg/L) after 72 hours SDS
SA	1:52	0.06	3.1	4.5	>0.035 (>58%)	<0.025
	1:15	0.21	3.1	5.0	0.16 (76%)	0.05
	1:8	0.39	3.1	5.0	0.29 (74%)	0.10
WA	1:13	0.55	7.2	7.5	0.47 (85%)	0.08
	1:8	0.9	7.2	7.5	0.8 (89%)	0.10
	1:4	1.8	7.2	7.5	1.44 (80%)	0.36

in our samples. Assuming that bromide, HOCl/OCl^- and DOC are in equilibrium, coupled with the fact that similar percentages of bromide were consumed in all Br:DOC samples, suggests that HOCl/OCl^- and DOC were not limiting the reaction. Rather, the amount of bromide incorporated appeared to be dependent on the initial bromide concentration not the Br:DOC ratio, although this could not be conclusively proved due to the limited data and difficulty of modifying the Br:DOC ratio without changing the bromide concentration.

Effect of bromide on DBPs

Comparison of DBP results across the two waters (Table 3) showed that increasing bromide resulted in increased concentration of THMs after 72 hours, with the total THM concentrations achieved for the highest bromide concentrations for each water source approximately double that at the initial/lowest bromide concentrations. This was caused by conversion to increased amounts of the brominated species. The extra bromide had less effect on the total HAA concentration, particularly for the WA water source. This is likely to have been a result of more substitution reactions occurring within the various HAA species rather than the creation of new HAAs.

As the bromide concentration increased, the total AOX concentration did not change appreciably, with the exception of the highest bromide concentration (1.8 mg/L, WA water source) where an increase from 707 μg/L (0.9 mg/L) to 985 μg/L (1.8 mg/L) was observed (approximately 43% increase) suggesting a stoichiometric barrier against greater formation had been overcome.

When evaluated as individual THM speciation, at all comparative time intervals throughout the SDS experiment,

Table 3 | Comparison of DBP species for SA and WA treated water with increasing bromide addition

SA treated water

Br concentration		0.06 mg/L			0.21 mg/L			0.39 mg/L		
DBPs		**0.5 hours**	**4 hours**	**72 hours**	**0.5 hours**	**4 hours**	**72 hours**	**0.5 hours**	**4 hours**	**72 hours**
THM species (μg/L)	Chloroform	10	18	57	6	12	40	4	7	20
	Bromodichloromethane	13	18	28	16	27	52	16	28	57
	Dibromochloromethane	5	7	9	19	28	40	30	45	75
	Bromoform	<1	<1	<1	5	6	7	18	20	25
	Total THMs (THM4)	**28**	**42**	**94**	**46**	**72**	**139**	**68**	**100**	**176**
HAA species (μg/L)	Chloro-HAAs	12	21	62	6	9	39	4	7	38
	Chloro-bromo-HAAs	11	17	32	18	32	58	21	37	79
	Bromo-HAAs	1	1	2	4	6	13	11	15	29
	Total HAAs (HAA9)	**24**	**39**	**96**	**28**	**47**	**110**	**36**	**59**	**146**
AOX (μg/L)		**169**	**199**	**287**	**116**	**162**	**270**	**107**	**164**	**280**

WA treated water

Br concentration		0.55 mg/L			0.9 mg/L			1.8 mg/L		
DBPs		**0.5 hours**	**4 hours**	**72 hours**	**0.5 hours**	**4 hours**	**72 hours**	**0.5 hours**	**4 hours**	**72 hours**
THM species (μg/L)	Chloroform	15	25	59	9	16	37	4	5	7
	Bromodichloromethane	42	64	99	41	64	113	27	38	56
	Dibromochloromethane	60	83	100	80	117	179	86	118	201
	Bromoform	24	27	26	58	67	81	152	187	310
	Total THMs (THM4)	**140**	**199**	**283**	**188**	**264**	**409**	**269**	**348**	**573**
HAA species (μg/L)	Chloro-HAAs	18	24	69	9	14	28	4	6	8
	Chloro-bromo-HAAs	55	76	117	56	82	121	48	63	90
	Bromo-HAAs	29	33	42	45	56	74	95	115	146
	Total HAAs (HAA9)	**102**	**133**	**228**	**110**	**152**	**223**	**147**	**184**	**244**
AOX (μg/L)		**383**	**493**	**707**	**405**	**471**	**707**	**442**	**559**	**975**

the effect of increased bromide concentration was a shift in equilibrium away from chloroform towards the brominated species: bromodichloromethane, dibromochloromethane and bromoform. With the exception of bromoform formation in the lowest bromide concentration of both water sources, the concentration of all THM analogues increased with time within each individual SDS experiment. The overall concentrations of THM and HAA species in the SA water source were less than half the concentration of the WA water source, which is attributable to the difference in DOC concentration.

Speciation of HAAs is more difficult to interpret due to the larger number of analogues, increasing substitution and the possible conversion between HAAs and THMs (Heller-Grossman *et al.* 1993). The mono-substituted analogues were either not detectable or present in very low concentrations for both waters and the speciation profiles were dominated by di- and tri-halide analogues (not shown).

Increased bromide concentration in the WA water resulted in a shift from chloro- and chloro-bromo-HAAs to the bromo-HAAs, whereas in the SA water samples the species shifted from the chloro-HAAs towards both chloro-bromo-HAAs and bromo-HAAs. Overall, similar to the THMs, the higher bromide concentrations shifted the equilibrium to favour the brominated analogues and increased with time.

Effect of bromide on toxicity

It should be noted that, in the discussion below, we are not addressing the absolute toxicity of the treated water samples. This is not possible because the samples needed to be concentrated so that waters which were compliant with health-based DBP guidelines (i.e. were by definition 'non-toxic') could be analysed in the bioassay. Rather than concentrate everything, including salts that could interfere with the bioassay, we chose to use a selective solid phase extraction method that inevitably lost volatile compounds such as the THMs and probably some haloacetonitriles and halonitromethanes. The THMs are not expected to be strongly cytotoxic in comparison with other DBPs (Plewa *et al.* 2008), but nevertheless the loss of volatile DBPs could reduce the total cytotoxicity detected. However, given that the sample toxicity will likely be due to a range of volatile and non-volatile DBPs and adding bromide will increase brominated-DBP speciation across all DBP classes, it is expected that changes in cytotoxicity quantified by the bioassay will *correlate* with changes in toxicity in the water samples. Thus, some general conclusions can be drawn regarding the effect of initial bromide concentration on DBP toxicity.

The effect of increased bromide had a considerable effect on the toxicity of the water (Figure 1). Increasing the bromide concentration from 0.06 mg/L to 0.39 mg/L caused a 4-fold increase in cytotoxicity for the SA water, and this was achieved within 0.5 hours of chlorination, with no subsequent change up to 72-hour contact time. This implies that elevated bromide levels will result in increased DBP formation and resultant toxicity. Similar results were observed for the WA water where the majority of the cytotoxic response was present after just 0.5 hours chlorine contact. This indicates that the most toxic DBP species will be formed within the WTP, prior to entering the distribution system.

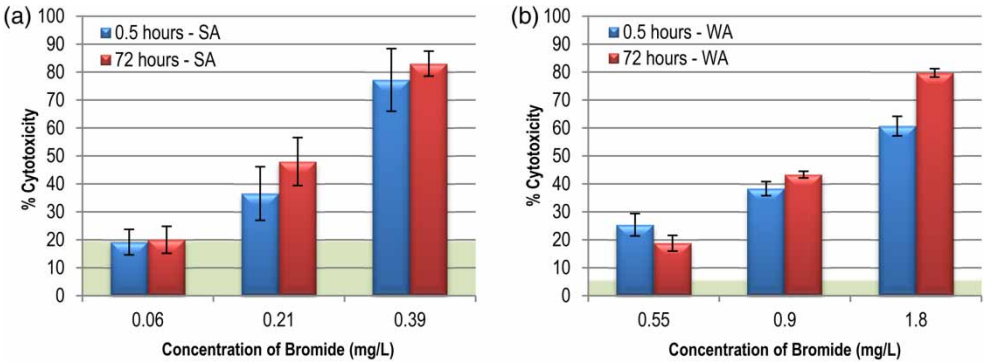

Figure 1 | Cytotoxicity comparison between 0.5 hours and 72 hours with respect to bromide concentration: (a) SA treated water; (b) WA treated water. Error bars are 1 standard deviation; shaded region represents negative control.

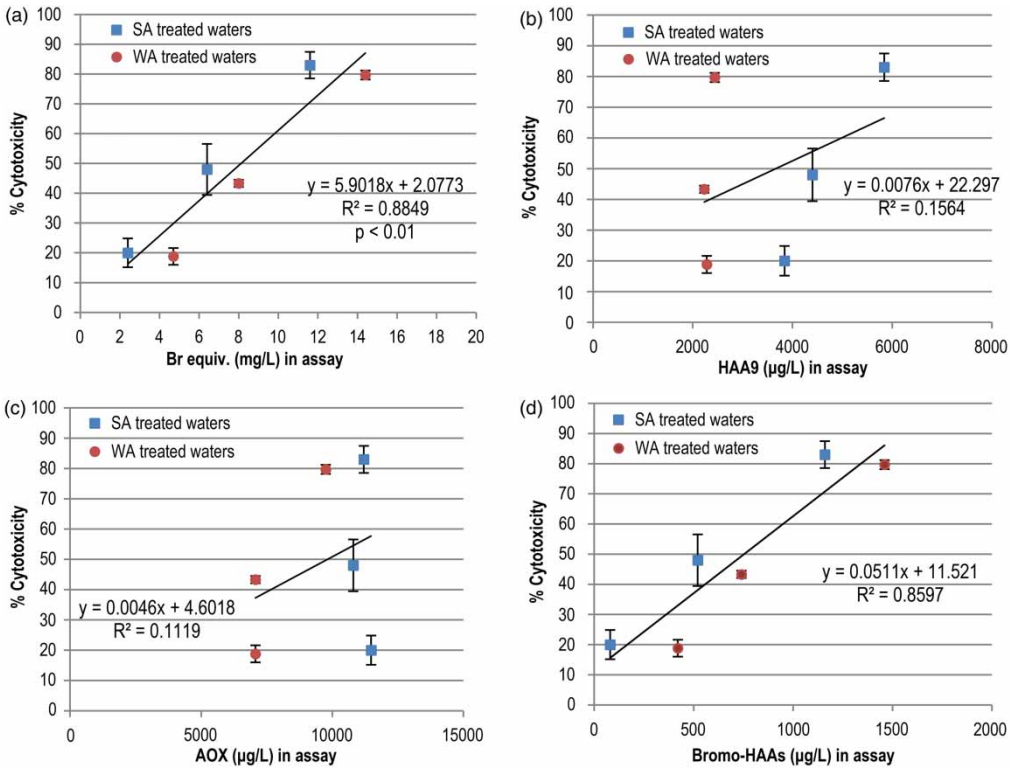

Figure 2 | Correlation of cytotoxicity after 72-hour SDS with: (a) Initial bromide concentration; (b) HAA9; (c) AOX; (d) bromo-HAAs. Error bars are 1 standard deviation.

In order to directly compare the results from the two waters, sample bromide, DOC (prior to chlorination) and DBP concentrations were multiplied by the concentration factors used in the cytotoxicity assays (SA water 40× and WA water 10×) to obtain the concentration equivalents present in the assays. The per cent cytotoxicity of each 72-hour sample was then plotted against the 'in assay' bromide equivalent concentration to investigate the relationship between bromide availability, HAAs and AOX and cytotoxicity (Figure 2(a)–(d)).

Cytotoxicity was linearly related to bromide availability irrespective of DOC concentration in chlorinated treated waters (Figure 2(a)) ($R^2 = 0.93$, regression $P < 0.01$, DOC equivalent SA treated 124 mg/L, WA treated 71.5 mg/L). Figure 2(b) and 2(c) show that cytotoxicity did not correlate with HAA9 or AOX concentrations, indicating they cannot be used as surrogates for cytotoxicity and their measurement is not appropriate to indicate the potential health impacts of DBP formation. However, cytotoxicity increased linearly with the three brominated HAAs ($R^2 = 0.86$), (Figure 2(d)), suggesting that brominated HAAs correlate with toxic species and may be partially responsible for the observed cytotoxicity. This would most likely be due to the presence of bromoacetic acid, the most toxic of the three brominated HAAs (Plewa *et al.* 2002), which was present in low concentration ($R^2 = 0.80$). Thus, the inclusion of the chlorinated HAAs in HAA9 may confound the results. It has been established that THMs are not present following the preconcentration step (Neale *et al.* 2012) and are of low potency compared to other DBPs (Plewa *et al.* 2008). Nevertheless, it was found that the THMs in the original chlorinated sample increased proportionally to the toxic compounds that were measured within the bioassay ($R^2 = 0.84$, not shown).

CONCLUSIONS

Treated waters from SA and WA water sources were spiked with additional bromide to achieve varying bromide to DOC ratios. The results for the two waters used in this study suggest that it is the concentrations of bromide and DOC present that determine bromide incorporation and DBP

speciation rather than the ratio (Br:DOC) itself, as the DOC concentration determines the chlorine dose and hence the Cl:Br ratio. Higher bromide concentrations increased total DBP concentration after 72-hour chlorine contact, although 100% of the bromide was not consumed.

Increasing bromide concentration resulted in increased amounts of THMs whilst total HAA and AOX remained relatively constant. Increased bromide shifted formation of measured DBPs towards the more bromine-substituted analogues.

A cytotoxicity assay confirmed that coagulation treatment significantly reduced the cell damage potential of chlorination while increasing bromide concentration increased it. Formation of cytotoxic products predominantly occurred within the first half hour, which would be while still within the WTP, and remained essentially constant for at least 72 hours, suggesting that these cytotoxic DBPs may be relatively stable once formed. A correlation was observed between cytotoxicity and the brominated HAAs suggesting that monitoring of brominated HAAs would provide an indication of potential health impacts. In addition, THMs also correlated with cytotoxicity, providing support for their use by regulators as surrogates for harmful DBPs.

In summary, if bromide concentrations in the water source increase, total DBP formation will increase, especially THMs, and there will be more brominated DBPs formed. Increased bromide will also result in more acute toxicity, with the majority of the cytotoxic response observed within the first half hour. With the limited knowledge regarding the specific causes of cytotoxicity, it is important for water utilities to continue to minimise potential precursors and to manage DBP formation to ensure safe drinking water for all consumers.

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