Cation Impact on N-Nitrosodimethylamine (NDMA) Formation from Ranitidine in Different Water Matrices

by

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A thesis submitted in conformity with the requirements for the degree of Master of Applied Science
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ABSTRACT

In previous research, ranitidine has formed high yields of the disinfection by-product N-Nitrosodimethylamine (NDMA) upon chloramination. In the current research, bench-scale experiments were conducted to investigate the impact of cations (i.e., Ca$^{2+}$, Mg$^{2+}$, and Na$^{+}$) on NDMA formation from ranitidine in three water matrices (Milli-Q® water, Lake Ontario water, and Otonabee River water) under practical chloramine disinfection conditions. In Milli-Q® water, excess cations did not change the yields of NDMA. NDMA formation kinetic profiles monitored in the lake and river water also indicated that elevating the cation concentrations did not affect the ultimate NDMA formation from ranitidine, but then did affect the observed rates of NDMA formation; the rates underwent an initial decrease and a subsequent increase as the cation concentrations were increased. The lowest reaction rates were observed in the lake and river water samples when they have a hardness level of 240 and 203 mg/L as CaCO$_3$, respectively.
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1 Introduction

1.1 Background

In order to maintain the water quality within distribution systems, secondary disinfection is required prior to water discharge. Chloramination has been recommended and popularly used in North America as a means of secondary disinfection mainly because of its longer-lasting treatment efficiency relative to chlorine (US Environmental Protection Agency (USEPA), 2006). A concern over adopting chloramination is the formation of N-Nitrosodimethylamine (NDMA) as a disinfection by-product (DBP). NDMA has a 2 to 4 orders of magnitude higher cancer risk than that of the typical DBPs (i.e., trihalomethanes and haloacetic acids) (Hrudey and Charrois, 2012), and it has been frequently detected in both finished water and distribution systems (MOE, 1998; CDHS, 2002; WHO, 2012). In order to lower the human cancer risk posed by NDMA via water consumption, the allowable NDMA concentration in drinking water has been regulated at 9 ng/L in Ontario (MOE, 2003) and 10 ng/L in Massachusetts (MassDEP, 2004) and California (OEHHA, 2006). Unlike the other DBPs, the formation of NDMA is rather slow (days instead of hours); therefore, more NDMA formation tends to take place in distribution systems and less in treatment plants (AWWA, 2006). Therefore, applying post treatment process (e.g., ultraviolet irradiation) to remove NDMA might not be sufficient to ensure the safety of water for household consumption; a more cost-effective way is to prevent its formation.

To date, NDMA formation has been mostly associated with the amines present in source waters. Most studies have focused on investigating NDMA conversion from model precursors (e.g., dimethylamine (DMA)). Only in recent years have pharmaceuticals been found to be potential precursors; some of them have exhibited high NDMA formation potential (FP) such as ranitidine. Ranitidine has shown an extraordinarily high capability to form NDMA, with molar conversions ranging from 60 to 90 % (Le Roux et al., 2011; Selbes et al., 2013; Shen and Andrews, 2011a). As a histamine H2-receptor antagonist, ranitidine is commonly prescribed for treating peptic ulcer diseases. Due to its frequent usage and improper disposal, it has been reported in different water sources, with concentrations ranging from 20 to 550 ng/L (Castiglioni et al., 2005; Zuccato et al., 2005; Batt et al., 2008). The mechanism of ranitidine to form NDMA upon chloramination has recently been proposed to involve a nucleophilic substitution process between monochloramine (NH₂Cl) and the dimethylamine group of ranitidine (Le Roux et al.,
The high yield has been suggested to be because the DMA group is attached to a benzyl-like functional group. In fact, tertiary amines with similar molecular structures have also been found capable of producing high yields of NDMA (Lee et al., 2007b; Sacher et al., 2008). Hence, efforts devoted to investigating the characteristics of ranitidine governing its formation of NDMA might also contribute to understanding the mechanisms of NDMA formation from other similar precursors.

A few studies have investigated the factors affecting NDMA formation from ranitidine using lab-grade water. The findings have indicated that the formation of NDMA is subject to some treatment operating conditions such as pH and chloramine species (Le Roux et al., 2012b; Selbes et al., 2013; Shen and Andrews, 2013a). In the very few studies involving real water matrices, the effect of water matrices has been found to significantly affect the observed NDMA formation rate, while barely affecting the ultimate yields; also, water with a higher total organic carbon (TOC) level seemed to result in a slower NDMA formation rate (Shen and Andrews, 2011b). The authors have suggested that the water matrix effect is primarily associated with the natural organic matter (NOM) present in water. It has been proposed that NOM tends to form reversible bonds with ranitidine, resulting in less ranitidine being available initially for reaction with chloramine, thus slowing down the observed rates of NDMA formation. However, the mechanism of such bonding has, as yet, remained uncertain. For example, other matrix components might be involved in the interactions between NOM and ranitidine, thus affecting the formation of NDMA. Therefore, to better characterize the water matrix effect, it is necessary to determine what influence, if any, other matrix components have on the NDMA formation from ranitidine.

So far, no study has been reported to investigate the impact of cations which occur naturally in water sources together with NOM. Given that cations have been proven to interact with NOM in different ways (e.g., charge neutralization and bridging), if the NOM-ranitidine binding is real, it is very likely that cations could interfere with such interactions. Since Canadian lakes and streams have shown a wide range of divalent cations (e.g., Ca$^{2+}$ and Mg$^{2+}$ at 2 to 1803 mg/L as CaCO$_3$) (Health Canada, 1979) and monovalent cations (e.g., Na$^+$ at less than 1 mg/L to more than 300 mg/L) (Health Canada, 1992), knowing the possible connection between the cation concentration and the NDMA formation pattern might help better evaluate the reported water matrix effects.
1.2 Research Objectives

The main objective was to investigate the impact of cation (i.e., Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^{+}\)) concentrations on the formation kinetics and the ultimate formation of NDMA from ranitidine.

1.3 Thesis Organization

Chapter 1 (i.e., this chapter) provides a very brief summary of the background for NDMA formation from ranitidine and states the research goals of this thesis.

Chapter 2 provides an overview of the existing knowledge about NDMA being a disinfection by-product, ranitidine as a potential precursor for NDMA, and the known factors affecting NDMA formation from ranitidine. It also identifies the research gaps that provided the rationale for conducting the current research.

Chapter 3 provides the materials and methods used in the experimental processes and data analysis.

Chapter 4 discusses the results of bench-scale experiments that explored the impact of cation concentrations on NDMA formation from ranitidine in Milli-Q\(^{®}\) water.

Chapter 5 discusses the results of bench-scale experiments that explored the impact of cation concentrations on NDMA formation from ranitidine in both Lake Ontario water and Otonabee River water.

Chapter 6 summarizes the conclusions from the current research and recommendations for future studies.
2 Literature Review

2.1 N-Nitrosodimethylamine (NDMA)

NDMA (Figure 2-1) belongs to a group of compounds called nitrosamines whose molecular structures usually comprise a tertiary amine and a nitroso group. NDMA can be found in manufacturing, consumables, and water systems. Like 90% of the other nitrosamines, NDMA is carcinogenic (Scanlan, 2000). Based on a 1 in 10^5 lifetime cancer risk, Health Canada has suggested a maximum acceptable concentration (MAC) for NDMA in drinking water at 0.04 µg/L (Health Canada, 2011). With the improvement of detection methods, trace amount (ng/L to µg/L) of nitrosamines have been reported in different water sources (California Department of Public Health (CDPH), 2011), leading them new targets in water related research. Among all the nitrosamines, NDMA has been of the greatest research interest because of its worldwide occurrence in various water sources.

![Figure 2-1. Molecular structure of NDMA](image)

2.1.1 NDMA as A Water Contaminant

NDMA as a wastewater contaminant was first found in a receiving surface water body of the industrial wastes released from a factory near Baltimore, Maryland where NDMA was used to produce unsymmetrical dimethylhydrazine (UDMH) (Fine et al., 1977). Industrial effluents like this usually contain high levels of NDMA, which could cause up to 1500 ng/L NDMA present in raw sewage (Mitch et al., 2003). Moreover, the low NDMA removal efficiency achieved by most of the conventional wastewater treatment processes has led to presence of NDMA (ng/L to µg/L) in the secondary effluents (Nawrocki and Andrzejewski, 2011; Sedlak et al., 2005). Although more than 70% of the NDMA precursors can be removed during biological treatments, considerable amount of them still appear in the secondary effluents (Sedlak et al., 2005). For water reuse and reclamation purpose, such effluents are often disinfected by chlorination or chloramination, which has been identified as an important source of NDMA; survey results have
linked most of these NDMA to the application of chloramination (Hatt et al., 2013; Krasner et al., 2009; Pehlivanoglu-Mantas and Sedlak, 2006b). More or less, the discharge of treated wastewater has led to the presence of NDMA and its precursors in the receiving water bodies (surface water or groundwater) (Nawrocki and Andrzejewski, 2011; Wang et al., 2012) which often are the source water for drinking water treatment plants, and as such, drinking water treatment plants located downstream are usually more vulnerable.

The concern for NDMA being a drinking water contaminant has been raised since 1989 when elevated amount (5 to 105 ng/L) of NDMA was detected in the finished water in Ohsweken, Ontario (Jobb et al., 1994). Afterwards, as high as 0.15 μg/L NDMA was detected in a drinking water well near a rocket engine testing facility in California in 1998 (CDPH, 2011). These incidents have prompted more monitoring for NDMA across North America, and progressively, NDMA has been detected in many drinking water systems (CDPH, 2011; Zhao et al., 2008). The occurrence of NDMA in drinking water has been found mostly associated with treatment plants using chloramine as secondary disinfectant. Among the nearly 1200 systems investigated in the Unregulated Contaminant Monitoring Rule 2 (UCMR-2) study by the US Environmental Protection Agency (USEPA), > 2 ng/L NDMA was observed in 34 % of the chloraminated systems, but only in 3 % of the chlorinated systems. In North America, NDMA concentration detected in the drinking water systems are generally below 10 ng/L with some exceptions (> 50 ng/L found in some chloraminated samples) (Russell et al., 2012). In contrast, limited occurrence of NDMA has been reported in Europe primarily due to the rare application of chloramination, except that 0.9 to 6 ng/L of NDMA has been found in Great Britain (Dillon et al., 2008; Goslan et al., 2009).

2.1.2 NDMA Formation Pathways in Drinking Water

NDMA formation is usually via reactions involving nitrogen-containing species. Several drinking water treatment processes have been found responsible for the formation of NDMA, such as chlorination, ozonation, activated carbon, and chloramination. A diagram of various NDMA formation pathways is given in Figure 2-2.
NDMA formation upon chlorination has been suggested to involve the formation of an \( \text{N}_2\text{O}_4 \) intermediate (Figure 2-2 C) which is subsequently converted to NDMA via nitrosation. Although this reaction is rapid, the low yield to NDMA has limited its contribution to the NDMA occurrence in drinking water systems. Yet this pathway can be important particularly when high amount of chlorine and nitrite co-occur (Krasner et al., 2013). The application of ozone could also lead to NDMA formation. For example, ozonation of the coagulant polyDADMAC could result in low yields of NDMA (Padhye et al., 2011). It has been noticed that ozonation could raise important concern over NDMA formation when it is applied to treat hydrazines or sulfamides impacted water. Scenarios as such have been reported in Japan and Germany (Kosaka et al., 2009; Schmidt and Brauch, 2008). Furthermore, applying ozone for water which contains > 500 µg/L bromide ions could also produce considerable amount of NDMA (Figure 2-2 D) (Shah and Mitch, 2012). In recent years, the application of activated carbon has also been identified as a possible source of the trace amount NDMA formation (Krasner et al., 2013).
The mechanism of NDMA formation upon chloramination has been proposed and later revised. The earliest version suggested that NDMA is formed via a nucleophilic substitution between monochloramine (NH$_2$Cl) and secondary amines, during which UDMH is formed as an intermediate (Figure 2-2 A). This mechanism has also been suggested for NOM being the precursor, under which circumstances, NOM is oxidized to release smaller precursors such as DMA (Chen and Valentine, 2006, 2007). However, given that the yields of NDMA from equivalent mole of UDMH and DMA are significantly different, this mechanism was questioned. Later studies have proposed another mechanism (Figure 2-2 B) in which dichloramine (NHCl$_2$) is responsible for the major formation of NDMA instead of NH$_2$Cl. Although NH$_2$Cl is the predominant chloramine species in chloramine disinfection, due to its disproportionation, NHCl$_2$ almost always coexist. The intermediate product, therefore, is the chlorinated UDMH (Cl-UDMH) which is subsequently oxidized either by oxygen to form NDMA or by chloramines to form other products (Chen and Young, 2008; Schreiber and Mitch, 2006). This revised mechanism has indicated the important role of the dissolved oxygen and NHCl$_2$, and more importantly, it is also applicable when pre-formed NH$_2$Cl is used (Shah and Mitch, 2012). It is worthwhile to mention that this mechanism is pH-dependent since pH affects the amine and chloramine speciation, the two predominant factors affecting the NDMA formation. A recent study has also suggested the important role of bromide ions in facilitating the formation of NDMA upon chloramination (Le Roux et al., 2012a), but the actual mechanism remain uncertain.

Among all the treatment processes, chloramination is potentially the most important one and accounts for most NDMA occurrences because of its relatively high yields to NDMA (Mitch et al., 2009; Boyd et al., 2011). In order to limit the formation of the typical disinfection by-products trihalomethanes (THMs) and haloacetic acids (HAAs) during chlorination, chloramine has been considered as an alternative residual disinfectant (Hubbs et al., 1981). In 2006, the USEPA has recommended chloramination as the best existing technology for consecutive systems owing to its effectiveness in reducing the formation of THMs and HAAs, while maintaining stable residuals with less taste and odor issues at a low installation and operating cost (USEPA, 2006). However, it did not take too long for people to realize that the practice of chloramination usually complies with the formation of NDMA as a DBP.
2.1.3 Strategies for NDMA Formation Control

Different guidelines or regulations for NDMA concentration have been advised by several organizations. Public health goals for NDMA in drinking water with respect to a $10^{-5}$ cancer risk have been recommended as 100 and 40 ng/L by World Health Organization (WHO) (2008) and Health Canada (2011), respectively. In addition, NDMA has been regulated in Ontario (9 ng/L; MOE, 2003), Massachusetts (10 ng/L; MassDEP, 2004), and California (10 ng/L; OEHHA, 2006). In particular, NDMA has been listed in the UCMR-2 as well as the drinking water contaminant candidate list 3 (CCL3) by the USEPA (USEPA, 2009). The USEPA is intended to make a preliminary decision on NDMA regulation in 2013, and will propose in 2016 if they decide to carry out the regulation process (Krasner et al., 2013). In order to comply with the guidelines or regulations, strategies for NDMA formation control in drinking water have been discussed in previous literatures; they have been reviewed and summarized into the following two categories.

2.1.3.1 Removal after Formation

The most technically feasible treatment for NDMA removal is ultraviolet (UV) irradiation (Nawrocki and Andrzejewski, 2011). In fact, it has been currently applied in some wastewater reclamation plants for that purpose. According to some literature, a UV dose of 1000 mJ/cm$^2$ can achieve a 90% reduction of NDMA (Mitch et al., 2009; Health Canada, 2011). However, the cost for UV operating at such a high dosage (typical UV dosage for virus inactivation is 40 mJ/cm$^2$) has restricted its practical application. Yet it has been designed for a drinking water treatment plant to remove the NDMA in its treatment influents (Swaim et al., 2008). Besides, advanced oxidation processes (AOPs) have also been studied for their effectiveness in NDMA removal. Results from bench-scale experiments have suggested that the degradation of NDMA by AOPs is largely dependent on the hydroxyl scavenging capacity of the raw water (Lee et al., 2007b). Treatment processes such as air stripping, activated carbon adsorption, and reverse osmosis (RO) membranes however have been found to achieve poor NDMA removal efficiency in the bench-scale tests (WHO, 2012). In fact, given the slow formation kinetics of NDMA, more NDMA formation tend to take place in the distribution systems and less in the treatment plants (American Water Works Association (AWWA), 2006); applying post treatments to
remove NDMA might not be sufficient to ensure the safety of water for household consumption. Hence, the more effective way should be to prevent NDMA formation.

2.1.3.2 Minimization of Formation

Two key elements should be looked upon when trying to minimize the NDMA formation: precursors and treatment operating conditions.

The ideal way of minimizing NDMA formation is to remove the precursors prior to secondary disinfection. The most direct approach is to avoid using source water that is characterized by high wastewater or industrial waste loads. Within the treatment, several processes have been found capable of removing NDMA precursors, such as conventional biological treatments (Pehlivanoglu-Mantas and Sedlak, 2006a), riverbank filtration (Krasner et al., 2012b), and physical powdered/granular activated carbon (PAC/GAC) adsorption (Hanigan et al., 2012; Sacher et al., 2008). Yet in the meantime, some treatment processes have been found to promote the NDMA formation potential (FP) by transforming the precursors into more potent forms. For example, ozonation of Polydiallyldimethylammonium (polyDADMAC) could transform the quaternary ammonium ring groups of polyDADMAC into a DMA which is more potent NDMA precursor (Padhye et al., 2011). Treatment processes have also found to serve as the sources for NDMA precursors. For example, coagulation and filtration have been found to significantly increase the NDMA FP, in which polymers (mostly amine-based) were added to improve their efficiency (Krasner et al., 2012a; 2013; Sedlak and Kavanaugh, 2006). Moreover, an increase in polymer dose has been found to result in a linear increase in NDMA formation (Kohut and Andrews, 2003). Thus, optimizing these treatment processes regarding to their use of polymer might help to minimize the NDMA formation.

Applying pre-oxidation methods prior to secondary disinfection have been found effective in inhibiting the organic NDMA precursors, such as free Cl₂ (Lee and von Gunten, 2010), O₃, O₃/H₂O₂, and ClO₂ (Lee et al., 2007a; Yang et al., 2013), ferrate (Lee et al., 2008; Yang et al., 2013), and K MnO₄ (Chen and Valentine, 2008). Since chlorine is commonly used in primary disinfection, it seems very cost-effective to apply pre-chlorination for NDMA precursor inhibition (Shah and Mitch, 2012). However, some evidence has suggested that pre-oxidation methods such as O₃ and free Cl₂ sometimes could enhance the NDMA FP of specific precursors (Padhye et al., 2011; Shen and Andrews, 2013). Other than the impact from pre-treatments, the
minimization of NDMA formation is also affected by the way to perform chloramination. As suggested by (Schreiber and Mitch, 2005), the best way to perform chloramination is to use pre-formed NH$_2$Cl. In fact, it is common practice to perform chloramination with inline-formed NH$_2$Cl by adding ammonia to pre-chlorinated water, which is considered an effective way for NDMA formation control (less NHCl$_2$ formed in producing NH$_2$Cl) (Schreiber and Mitch, 2005).

In practice, the formation of NDMA is to a great extent affected by several treatment operational parameters: pH, NH$_2$Cl dosage, and contact time. pH affects both the chloramine species and the NDMA precursor forms (Shen and Andrews, 2013a). For example, a high pH on the one hand would lead to less disproportionation of NH$_2$Cl, and on the other hand, it would result in more non-protonated form of precursors; the two outcomes affect the NDMA formation oppositely. Crittenden et al. (2012) observed a peak NDMA formation at pH 7 in the raw water collected from the West Branch of the California State Aqueduct. It has been suggested that an elevated or a reduced pH could inhibit the NDMA formation (Najm and Trussell, 2001; Sedlak and Kavanaugh, 2006; Sacher et al., 2008). However, it should be noticed that the optimal pH regarding NDMA formation control should be case-specific.

Najm and Trussell (2001) found a rise in NDMA formation as the chloramine dosage was increased. There has been discussion about moderately adjusting the disinfectant dosage to reduce DBP formation without compromising the disinfection sufficiency (Crittenden et al., 2012). Except for lowering the disinfect dosage, reducing the contact time is also a means of minimizing NDMA formation. The total contact time for NDMA formation is comprised of two parts: the in-plant and the off-plant. The in-plant contact time refers to the water storage time inside the treatment plant (usually in hours), while the off-plant contact time refers to the water retention time in distribution systems (usually in days). The results of a survey of more than 800 U.S. utilities indicated an average distribution system retention time of 1.3 days and a maximum retention time of 3.0 days. Since the retention in distribution system is largely dependent on the actual water consumption, a more feasible way to shorten the NDMA formation contact time is to adjust the in-plant retention time. Krasner et al. (2012b) have found a decrease in NDMA formation by reducing the in-plant contact time from half day to a few hours. However, due to the slow formation kinetics of NDMA, instead of forming in plant, a large portion of NDMA is expected to form within the distribution systems.
2.2 NDMA Precursors

2.2.1 NDMA Precursors in General

Amines and amides are the typical NDMA precursors. Particularly, secondary amines (e.g., DMA) have been frequently studied as model precursors (Choi and Valentine, 2002; Schreiber and Mitch, 2005; Shah and Mitch, 2012); they react with chloramine directly forming NDMA in a short time. In contrast, amides forming NDMA has been found orders of magnitude slower since its nitrogen atom is adjacent to the electron-withdrawing carbonyl groups (Mitch and Sedlak, 2004). It has been suggested in general that, as a precursor reacts with chloramine, a secondary amine, which began as the amine group of the precursor, is released to react with chloramine forming NDMA. Therefore, for most of the tertiary and quaternary amines, their conversion rates to NDMA are similar to that of the secondary amines. Exceptions have been found for certain tertiary amines (e.g., ranitidine) of which the nitrogen β-position is attached to an aromatic group; they have been found to exhibit high conversions to NDMA (Le Roux et al., 2011, Shen and Andrews, 2011a).

NOM (Chen and Valentine, 2008), bulk dissolved organic nitrogen (DON) (Chang et al., 2011), soluble microbial products (SMPs) (Krasner et al., 2008), and pharmaceuticals and personal care products (PPCPs) (Kemper et al., 2010; Shen and Andrews, 2011a) have also been proved to have certain levels of NDMA FP. Recently, studies have indicated a new source of NDMA formation, which is the synthetic materials that are used in the distribution systems (Morran et al., 2011). Regarding the NDMA precursors, Krasner et al. (2013) have summarized their relative importance for NDMA formation control in drinking water treatments in Table 2-1 based on the current state of knowledge.
Table 2-1. Relative importance of different NDMA precursors in drinking water perspective (reproduced from Krasner et al., 2013)

<table>
<thead>
<tr>
<th>Precursor</th>
<th>Importance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amine-based coagulation polymers</td>
<td>High</td>
</tr>
<tr>
<td>Effluent-impacted source waters</td>
<td>High</td>
</tr>
<tr>
<td>Pharmaceuticals and personal care products</td>
<td>Moderate/High</td>
</tr>
<tr>
<td>Distribution system materials</td>
<td>Moderate</td>
</tr>
<tr>
<td>Anion exchange resins</td>
<td>Moderate</td>
</tr>
<tr>
<td>Soluble microbial products</td>
<td>Limited</td>
</tr>
<tr>
<td>Agricultural chemicals</td>
<td>Limited</td>
</tr>
<tr>
<td>Bulk DON</td>
<td>Low</td>
</tr>
<tr>
<td>Algae</td>
<td>Low</td>
</tr>
</tbody>
</table>

As listed above, because of their frequent occurrence in water sources and potential to produce high yields of NDMA, pharmaceuticals as precursors have been considered as an important source of NDMA formation. The concern has grown as an increasing variety of pharmaceuticals have progressively been found in water sources (Chang et al., 2011; Blair et al., 2013).

2.2.2 Ranitidine

Ranitidine (trade name Zantac®), as a histamine H2-receptor antagonist, is commonly prescribed for treating peptic ulcer diseases. It was first introduced in 1981, and became the world’s biggest-selling prescription drug in the following decade (Wright, 1996). Although nowadays, it has been progressively replaced by proton pump inhibitors, it still remains in the top 15 sold-list of prescribed drugs around the world (Fent et al., 2006). However, the frequent usage of ranitidine has led to its presence in water systems. Because of its large molecular size and low polarity, ranitidine has been inefficiently removed by most of the conventional treatment processes (e.g., activated sludge, pre-chlorination) (Krauss et al., 2010). In recent years, ranitidine has been studied as a NDMA precursor because of its extraordinarily high yield (60 to 90 %; Le Roux et al., 2011; Sacher et al., 2008; Shen and Andrews, 2011a) to NDMA.
2.2.3 Ranitidine Occurrence and Removal

Medical wastes discharge usually contains pharmaceuticals at high concentrations: Gómez et al. (2006) found 400 to 1700 ng/L of ranitidine in a hospital effluent in Spain. Ranitidine has also been found in raw sewage mainly due to its improper disposal (Godfrey et al., 2007). Owing to its extensive use, ranitidine has been detected in municipal wastewater. For example, up to 758 ng/L of ranitidine were found in several sewages in Western Balkan region (Bosnia and Herzegovina, Croatia and Serbia) (Terzić et al., 2008); 20 to 550 ng/L of it has been reported in Italy and the U.S. (Castiglioni et al., 2005; Zuccato et al., 2005; Batt et al., 2008). In wastewater impacted surface water, ranitidine has also been detected, but in lower concentrations. Zuccato et al., (2000) found up to 10 ng/L of ranitidine in the rivers in Italy, and a higher concentration was detected in the sediments. A survey results have indicated around 0.01 µg/L ranitidine existed in 1.2 % of the U.S. streams (Kolpin et al., 2002).

Water treatment processes have been tested for ranitidine removal efficiency. UV photolysis was able to degrade ranitidine, the efficiency of which could be enhanced by adding TiO₂ as photocatalyst (Addamo et al., 2005). Conventional activated sludge achieved a low NDMA removal rate (17 to 26 %) (Carucci et al. 2006) which is similar to that of ozonation (20 to 25 %) (Rivas et al. 2009); a higher removal of ranitidine could be attained by membrane bioreactors (MBRs) (Radjenović et al., 2009). Recently, advanced treatment processes have been found to achieve excellent removal efficiency of ranitidine: Photosensitized oxidation performed a nearly complete removal of ranitidine within 30 min (Lee et al., 2011); a combination of membrane bioreactor coupled with reverse osmosis (MBR-RO) achieved up to 95 % ranitidine removal in a pilot-scale test (Dolar et al., 2012); similar efficiency was obtained by a nanofiltration system (López-Muñoz et al., 2012). However, ranitidine removal has been mainly referred to its degradation; NDMA would still be formed if the amine moieties are not completely mineralized.

2.3 NDMA Formation from Ranitidine upon Chloramination

2.3.1 Formation Pathways

Ranitidine has a high yield to NDMA during chloramine disinfection: Its molar conversion was found as 80.2 % (Le Roux et al., 2011), 80.5 % (Selbes et al., 2013), 82.7 % (Shen and Andrews, 2011b), and 89.9 % (Shen and Andrews, 2011a) in recent studies. It has been suggested that...
NDMA formation from tertiary amines involves a chlorine transfer process in which DMA is released and yields NDMA afterwards (Mitch and Schreiber, 2008). However, if a chlorine transfer occurs on ranitidine, the released moieties (DMA or dimethylchloramine) could only yield $<3\%$ of NDMA (Choi and Valentine, 2002; Mitch and Sedlak, 2001, 2004), which is not comparable to the high yields from ranitidine. By looking into the chloraminated by-products of ranitidine, Le Roux et al. (2012b) have proposed a different formation pathway which involves a nucleophilic substitution process. The detailed mechanism is given in Figure 2-3.
Figure 2-3. NDMA formation pathway from ranitidine upon chloramination proposed by Le Roux et al. (2012b)
This mechanism suggested a fast chlorine transfer from NH$_2$Cl to ranitidine forming chlorinated ranitidine analogues; chlorine atom would preferentially attack the thioethyl-N-methyl-2-nitroethene-1,1-diamine moiety and the sulfur atom. The attack on the sulfur atom could either form sulfoxide compounds or cause the cleavage of the C-S bond, the latter of which would lead to a release of 5-(dimethylaminomethyl)furfuryl alcohol (DFUR). The release of DFUR is a substantial step in this mechanism. DFUR has a > 50 % molar yield to NDMA (Sacher et al., 2008), which was suggested to contribute nearly the overall NDMA formation from ranitidine (Le Roux et al., 2012b). In this mechanism, NDMA is formed directly via a neucleophic substitution between NH$_2$Cl and the DMA moiety from the released DFUR or the chlorinated ranitidine analogues. There are several factors have been found affecting the NDMA formation from ranitidine; they have been summarized in the following contents.

2.3.2 The Effect of Molecular Structure

The high NDMA conversion rate from ranitidine has been attributed to its molecular structure. Lee et al., (2007b) and Sacher et al. (2008) suggested that tertiary amines whose DMA group connected to a benzyl-like functional group (ring structure) are capable of yielding high amount of NDMA. Furthermore, it has been suggested that higher conversion to NDMA could be achieved if there is only one carbon between the ring structure and the DMA group (Sacher et al., 2008); Selbes et al. (2013) have linked the high NDMA FP of such amines to the release of a stable leaving group: when a nucleophilic substitution occurs on the DMA moiety, such amines would tend to release a carbocation which is largely stabilized by resonance whether or not the heteroatoms (e.g., O, N, and S) are present in the rings. As for ranitidine (Figure 2-3), when forming NDMA, its leaving group is a carbocation which is attached to the benzylic position of a furan ring; such carbocation is thermodynamically stable, thus facilitating the NDMA formation. Besides, the electron density of the leaving group also matters. Shen and Andrews (2011a) examined the NDMA FP of 20 pharmaceuticals among which those with their DMA group bonded to an electron-rich leaving group were found to have higher yields to NDMA. Similarly, it has been suggested that the oxygen atom in the furan ring of ranitidine could enhance the electron density of the leaving group, which has facilitated the NDMA formation (Selbes et al., 2013)
2.3.3 The Effect of pH

pH impact on ranitidine to form NDMA has been studied mostly in Milli-Q® water samples. Le Roux et al. (2011) tested the NDMA formation from ranitidine under pH ranging from 4 to 10, and observed the peak formation at pH 7.9. In agreement with it, Shen and Andrews (2013a) have suggested the optimal pH range for ranitidine to form NDMA is from 7 to 8. The authors suggested that a basic environment (i.e., pH>8) would result in the lack of dichloramine (a chloramine species distribution diagram under different pH condition is given in Figure 2-4), thus inhibiting NDMA formation.

![Distribution diagram of chloramine species under different pH values](image)

**Figure 2-4. Distribution diagram of chloramine species under different pH values (from USEPA, 1999)**

For the NDMA formation in acidic environment (i.e., pH<7), with respect to the two mechanisms mentioned in 2.3.1, Shen and Andrews (2013a) gave two possible explanations. If the electrophilic substitution mechanism proposed by Mitch and Schreiber (2008) is true, non-protonated form of precursor would be favored in forming NDMA; therefore, the acidic pH resulting in protonated ranitidine being the dominant species would inhibit the NDMA formation. If the nucleophilic substitution mechanism (Figure 2-3) is true, the acidic pH might inhibit the acid release step (e.g., NH- reacts with dissolved oxygen), thus reducing the ultimate
NDMA formation. Besides the pH impact on the ultimate NDMA formation, Shen and Andrews (2013a) also pointed out the pH impact on NDMA formation kinetics: a higher pH could enhance the observed reaction rates. Since in either mechanism, the reaction of forming NDMA occurs mainly between chloramine and the non-protonated DMA moiety, a higher pH would favor the formation of NDMA by providing more non-protonated ranitidine.

2.3.4 The Effect of Chloramine Speciation

Monochloramine is preferably used in chloramination because it causes less taste and odor issues comparing with dichloramine and nitrogen trichloride (USEPA, 1999). A theoretical breakpoint chlorination curve is given in Figure 2-5. Monochloramine is preferentially formed with a Cl\(_2\):NH\(_4\)-N ratio (by weight) < 5:1 under typical water treatment pH (6.5 to 8.5).

![Figure 2-5. Theoretical breakpoint chlorination curve (from (USEPA, 1999))](image)

In order to ensure NH\(_2\)Cl is the only chloramine species, meanwhile preventing issues caused by excess ammonia (e.g., nitrification and biofilm growth), common practice is to control the Cl\(_2\):N ratio in the range of 3:1 to 5:1 with the optimal as 4:1 (USEPA, 1999). However, due to the slow degradation of NH\(_2\)Cl, without modification of pH or Cl\(_2\):N ratio, NHCl\(_2\) often coexist (43 % NH\(_2\)Cl to 57 % NHCl\(_2\)) (USEPA, 1999). It has been reported that a small fraction of dichloramine could significantly enhance the NDMA formation from tertiary amines (Mitch et al., 2005). Similarly, increasing the Cl\(_2\):N ratio has resulted in a higher NDMA formation from a mixture of pharmaceutical-based NDMA precursors (Shen and Andrews, 2011a). Surprisingly, Selbes et al. (2013) found dichloramine had little impact on NDMA formation form ranitidine:
ranitidine forming NDMA was performed with and without excess ammonia, and negligible difference was observed in NDMA formation after 5 day incubation. The presence of excess ammonia has been suggested capable of suppressing the degradation of monochloramine to dichloramine (Hong et al., 2007). However, as suggested by Selbes et al. (2013), the dichloramine in samples with excess ammonia was measured as below the detection limit (0.05 mg/L); in another word, there was no guarantee that dichloramine was absolutely absent. Therefore, further investigations are needed to confirm whether the presence of dichloramine would affect the NDMA formation from ranitidine.

### 2.3.5 The Effect of NOM

Two NOM fractions (Myrtle Beach transphilic and Myrtle Beach hydrophobic) were tested for their impact on NDMA formation from ranitidine; they were found to significantly enhance the yields of NDMA (Selbes et al., 2013). The results have indicated that certain fractions of NOM could increase the NDMA formation, yet given that NOM comprises a complex variety of fractions in real water scenario, real water matrix effect could be different. Shen and Andrews (2011b) have studied the NDMA formation from ranitidine in real water matrix samples which contained different levels of NOM. The real water matrices were found to slow down the observed rates of NDMA formation and had little impact on the ultimate formation; the authors have attributed the impact to the presence of NOM: since NOM is mostly negatively charged and ranitidine is positively charged at neutral pH, the slower formation was suggested as a result of the electrostatic attraction between NOM and ranitidine. The findings have also indicated that a higher total organic carbon (TOC) level could result in a slower formation of NDMA from ranitidine.

### 2.4 Research Gap and Hypotheses

To date, most of the studies regarding NDMA formation from ranitidine were done in lab-grade waters, and very few have introduced real water matrices. In the studies involving real water samples, the matrix effect has been mainly associated with the interactions between NOM and ranitidine (Shen and Andrews 2011b). Given that the mechanism of such interactions is as yet remained uncertain, it is possible that other water matrix components are also involved. Moreover, it has been hypothesized that the interactions are through electrostatic attraction;
therefore, theoretically, any compound with charges should have the potential to affect the binding, thus affecting NDMA formation from ranitidine in the presence of NOM.

Cations with positive charges occur naturally in real waters, making them good targets to look into; they have been found to affect both the aggregation and the dissolution processes of NOM (Brigante et al., 2009; Kloster et al., 2013). Meanwhile, divalent cations (i.e., Ca$^{2+}$, Mg$^{2+}$) which are often measured as hardness have been reported to be present at a broad range (2 to 1803 mg/L as CaCO$_3$) in Ontario lakes and streams (Health Canada, 1979). Therefore, this study examines the impact of cations on NDMA formation from ranitidine in different water matrices. A preliminary hypothesis has been proposed: cations with larger surface charge than protonated ranitidine, would preferentially take up the binding sites of NOM, thus resulting in less NOM-ranitidine binding; therefore, faster formation of NDMA from ranitidine should be expected; however, cations are hypothesized to have limited impact on the ultimate NDMA formation.
3 Materials and Methods

3.1 Materials

3.1.1 Chemicals and Working Solutions

All the chemicals used in this research were purchased from Sigma-Aldrich Canada (Oakville, Ontario). Ranitidine stock solution (0.1 M) was prepared by dissolving 0.35 g of ranitidine hydrochloride (C\textsubscript{13}H\textsubscript{22}N\textsubscript{4}O\textsubscript{3}S \cdot HCl) into 10 mL methanol and stored at 4 °C for use. The chlorine stock solution was prepared by diluting the 10 to 15 % (10000 to 15000 mg/L as Cl\textsubscript{2}) sodium hypochlorite (NaClO) by a factor of 10 and stored at 4 °C; the concentration was measured before use. Monochloramine working solution was prepared freshly by mixing ammonium chloride stock solution (3.5 g/L) and chlorine stock solution at a Cl\textsubscript{2}:NH\textsubscript{4}-N mass ratio of 4.2:1 and equilibrating for at least half an hour prior to use. Monochloramine concentration in the working solution was measured before use to ensure that the monochloramine to total chlorine ratio was more than 90 %.

In order to calculate the NDMA formation in samples, deuterated N-Nitrosodimethylamine (d\textsubscript{6}-NDMA, 98 % atom %D) and NDMA were used as the internal standard and reagent standard, respectively. The stock solutions of d\textsubscript{6}-NDMA (1 mg/L) and NDMA (1 and 10 mg/L) were all made by diluting the liquid reagents with methanol and stored at 4 °C for use. Lewatit\textsuperscript{®} AF 5 beads (Sigma-Aldrich) were used to extract both d\textsubscript{6}-NDMA and NDMA from the water samples; they were conditioned in a muffle furnace at 320 °C for 3 h and cooled down in a desiccator before use.

3.1.2 Water Matrices

In this research, three types of water were investigated: Milli-Q\textsuperscript{®} water, Lake Ontario water, and Otonabee River water. Milli-Q\textsuperscript{®} water was generated from a Millipore Reference Ultrapure Water Purification System (Fisher Scientific). Lake Ontario water was collected from the analytical lab in Lorne Park Water Treatment Plant (Mississauga, Ontario). Otonabee River water was collected from the intake of the Peterborough Water Treatment Plant (Peterborough, Ontario). Water quality parameters (i.e., pH, ultraviolet absorbance at 254 (UV\textsubscript{254}), total organic carbon (TOC), alkalinity) were measured after the water was transferred back to the lab at University of Toronto (Toronto, Ontario).
In this research, three cations (i.e., Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^+\)) were investigated for their impact on NDMA formation from ranitidine in the three selected water types. Ca\(^{2+}\) and Mg\(^{2+}\) are typical divalent cations existing in various types of water resulting in “hard water”. Sodium is an essential element to all animals and some plants, therefore naturally occur in water environment as well. Due to the fact that these three cations usually coexist in natural water bodies, they were investigated in this research as a group instead of individuals. The initial concentration of these cations in the selected water types are given in Table 3-1.

### Table 3-1. Initial cation concentrations in the three selected waters

<table>
<thead>
<tr>
<th></th>
<th>Milli-Q(^{®}) water(^a)</th>
<th>Lake Ontario water(^b)</th>
<th>Otonabee River water(^c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca(^{2+}) (mg/L)</td>
<td>N/A</td>
<td>42.3</td>
<td>39.3</td>
</tr>
<tr>
<td>Mg(^{2+}) (mg/L)</td>
<td>N/A</td>
<td>10.1</td>
<td>3.1</td>
</tr>
<tr>
<td>Na(^+) (mg/L)</td>
<td>N/A</td>
<td>10.4</td>
<td>4.7</td>
</tr>
</tbody>
</table>

\(^a\) Ionic traces in Milli-Q\(^{®}\) water were not detected  
\(^b\) Sadmani et al., 2014  
\(^c\) Sadmani, 2013

Cations were introduced to the three selected water types as a combination of Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^+\). By using “X” to represent a combination of 40 mg/L of Ca\(^{2+}\), 20 mg/L of Mg\(^{2+}\), and 10 mg/L of Na\(^+\), the addition dosages could be described as ratios of X. In total, there were six addition dosages applied in this research, as summarized in Table 3-2.

### Table 3-2. Cation addition dosages applied in the experiments

<table>
<thead>
<tr>
<th></th>
<th>0X (no addition)</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
<th>5X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca(^{2+}) (mg/L)</td>
<td>0</td>
<td>10</td>
<td>20</td>
<td>40</td>
<td>80</td>
<td>160</td>
<td>200</td>
</tr>
<tr>
<td>Mg(^{2+}) (mg/L)</td>
<td>0</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>40</td>
<td>80</td>
<td>100</td>
</tr>
<tr>
<td>Na(^+) (mg/L)</td>
<td>0</td>
<td>2.5</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>40</td>
<td>50</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca\(^{2+}\), 20 mg/L Mg\(^{2+}\), and 10 mg/L Na\(^+\) of added cations

The addition of Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^+\) was performed by dosing CaCl\(_2\), MgSO\(_4\), and NaCl stock solutions; CaCl\(_2\) (500 g/L; 180.2 g/L as Ca\(^{2+}\)), MgSO\(_4\) (100 g/L; 20.0 g/L as Mg\(^{2+}\)) and NaCl (100 g/L; 39.3 g/L as Na\(^+\)) stock solutions were prepared by dissolving calcium chloride dihydate, magnesium sulfate, and sodium chloride in Milli-Q\(^{®}\) water, respectively.
3.1.3 Glassware

All the glassware and vials were purchased from VWR Canada (Mississauga, Ontario). 1 L amber bottles with low density polyethylene caps were used to perform the NDMA formation tests. They were made chlorine-demand-free after purchase: the bottles were washed and rinsed thoroughly, dosed with 2 mL of 10 to 15 % (10000 to 15000 mg/L as Cl₂) sodium hypochlorite, and filled with deionized (DI) water to be head-space free; the bottles were emptied after 24 h, rinsed thoroughly with distilled water, and air dried. Before each use, they were washed in a dishwasher, rinsed three times with distilled water, and dried in the oven at 250 °C for 6 h.

3.2 Analytical Methods

3.2.1 Water Quality and Chloramine Measurements

Analytical methods for water quality parameters and chloramine related measurements are summarized in Table 3-3. Samples were measured in duplicate for each parameter.

Table 3-3. Analytical methods for water quality parameters and chloramine related measurements

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Unit</th>
<th>Instrument</th>
<th>Reference Method*</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>N/A</td>
<td>Model 8015 pH meter/VWR Scientific Inc., Mississauga, Ontario</td>
<td>N/A</td>
</tr>
<tr>
<td>TOC</td>
<td>mg/L</td>
<td>Aurora 1030 TOC analyzer/O.I. Analytical, College Station, Texas</td>
<td>SM 5310C</td>
</tr>
<tr>
<td>Alkalinity</td>
<td>mg/L as CaCO₃</td>
<td>N/A</td>
<td>SM 2320B</td>
</tr>
<tr>
<td>UV₂₅₄</td>
<td>cm⁻¹</td>
<td>CE3055 Reflectance Spectrophotometer/ Cecil Instruments Ltd., Cambridge, England</td>
<td>SM 5910B</td>
</tr>
<tr>
<td>Total chlorine</td>
<td>mg/L</td>
<td>DR 2800 spectrophotometer HACH® kit</td>
<td>4500-ClF</td>
</tr>
<tr>
<td>Monochloramine</td>
<td>mg/L</td>
<td>DR 2800 spectrophotometer HACH® kit</td>
<td>4500-ClF</td>
</tr>
</tbody>
</table>

* From EPA Standard Method (APHA-AWWA-WEF, 2005); N/A stands for not applicable

3.2.2 Liquid Chromatography-Organic Carbon Detection (LC-OCD) for NOM Fraction Analysis

NOM fractions in Lake Ontario water and Otonabee River water were analyzed using the LC-OCD analyzer at the University of Waterloo (Ontario, Canada). Duplicate LC-OCD samples
were prepared by filtering the raw water through a 0.45 µm membrane filter paper (Supur®-450 PES membrane filter, non-sterile) and shipped to the University of Waterloo for analysis within one week. LC-OCD system has been described in detail by Huber et al. (2011). Proprietary software (ChromCalc, DOCLABOR, Karlsruhe, Germany) was used for data acquisition and processing. The LC-OCD uses a size-exclusion column (SEC) to separate NOM fractions. Samples injected into the LC-OCD were separated into two portions: one went through a bypass stream diverted around the SEC column for the total dissolved organic carbon (DOC) analysis, while the other went through the SEC column for the hydrophilic DOC fraction analysis. The hydrophobic DOC fraction was calculated by subtracting the hydrophilic DOC from the total DOC since it could not be eluted through the SEC column within the limited measuring time. The hydrophilic DOC can be categorized into five main fractions; they were eluted in the order of biopolymers, humic substances, building blocks, low molecular weight (LMW) acids, and LMW neutrals. Before integrating the peaks, the boundary of each peak was assigned manually since complete separation of each individual peak cannot be achieved, and therefore some quantification variance might be introduced to the results. Such quantification accuracy varies among different fractions. In general, high accuracy (i.e., ± 2 to 7 %) could be achieved for fractions with high molecular weight (i.e., biopolymers and humic substances).

3.2.3 NDMA Analysis

NDMA concentrations were measured using Gas Chromatography-Mass Spectrometry (GC-MS) after being extracted from the water samples and concentrated into an organic solvent (dichloromethane (DCM)). An aliquot of 500 mL of sample was transferred to a clean 1 L amber bottle and spiked with 50 ng/L of d₆-NDMA as a surrogate to correct for variations in extraction efficiency. 200 mg of Lewatit® AF 5 beads were added to extract the d₆-NDMA and NDMA. The bottles were then capped with Teflon-lined caps and put on an orbital shaker (Thermolyne Bigger Bill M49235, Barnstead International, Asheville, N.C., USA) at 250 rpm for 1 h. Afterwards, the beads were filtered onto a filter paper (Whatman grade 4) which was later transferred to an aluminum tray and put in the fume hood to air dry for 30 min. Finally, the beads were transferred to a 2.0 mL GC auto-sampler vial and air dried for 2 h. The NDMA and d₆-NDMA extracted by the beads were eluted by adding 500 µL of DCM (99.9 %) into the vial. The vials were subsequently capped with Teflon-lined caps and ready for GC-MS analysis. As
suggested by Munch and Bassett (2004), the samples in these vials could be stored for up to 28 days at -15 °C or less.

Sample vials were put on a CombiPAL auto-sampler and the extracts were analyzed using a Varian 3800 GC coupled with a Varian 4000 ion trap MS (Agilent Technologies). A Varian 1079 injector fitted with a Carbofrit liner (Chromatographic Specialties; 3.4 mm ID, 5.0 mm OD, 54 mm length) and a programmed temperature vaporizer was used. A DB1701 (30 m x 0.25 mm x 0.25 μm) column was used. Chemical ionization (CI) was employed with high purity methanol as the reagent liquid. The GC-MS operating conditions for NDMA analysis are given in Table 3-4.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Injection volume</td>
<td>8 μL</td>
</tr>
<tr>
<td>Inlet temperature program</td>
<td>initially stayed at 25 °C for 0.8 min; increased by 200 °C/min to 240 °C and held for 24 min</td>
</tr>
<tr>
<td>Inlet split ratio</td>
<td>initially on (5); 0.8 min off; 6 min on (100)</td>
</tr>
<tr>
<td>Oven temperature program</td>
<td>initially held at 35 °C for 5.5 min; increased by 15 °C/min to 200 °C; increased by 40 °C/min to 240 °C and held for 10 min</td>
</tr>
<tr>
<td>Column flow</td>
<td>1.2 mL/min</td>
</tr>
<tr>
<td>Pressure pulse</td>
<td>stayed at 20 psi for 4 min</td>
</tr>
<tr>
<td>Scans Averaged</td>
<td>3 μScan</td>
</tr>
<tr>
<td>Emission current</td>
<td>50 μAmps</td>
</tr>
<tr>
<td>Electron Multiplier offset</td>
<td>+300 V</td>
</tr>
<tr>
<td>Filament delay</td>
<td>8.2 min</td>
</tr>
<tr>
<td>Ion indicator of NDMA</td>
<td>75 amu</td>
</tr>
<tr>
<td>Ion indicator of d₆-NDMA</td>
<td>81 amu</td>
</tr>
</tbody>
</table>
3.3 Experimental Protocol

3.3.1 Chloramine Dosage Determination

For the secondary disinfection, it has been regulated in Ontario that the chloramine concentration in a distribution system must be within the range of 0.25 to 3.0 mg/L, with a recommended concentration of 1 mg/L (MOE, 2006). In order to comply with the regulation as well as provide sufficient chloramine to react with ranitidine to form NDMA, the chloramine dosage applied in this research was selected to be 2.5 mg/L above the 24 h demand for the respective water matrix. Since the TOC level in Milli-Q® water has been suggested to be lower than 5 µg/L (EMD Millipore, 2013), the 24 h demand in Milli-Q® water was considered minimal (corroborated during the testing described in Chapter 4). Accordingly, the 24h chloramine demand test was performed only in Lake Ontario water and Otonabee River water.

For the 24 h chlorine demand tests, three initial monochloramine dosages (2.5, 5, and 10 mg/L as total Cl₂) were applied to each water type, and samples were prepared in duplicate. The total chlorine concentration in each sample was measured after 24 h (24 h total chlorine residual). A linear regression was applied between the initial chloramine dosage and the 24 h total chlorine residual. The chloramine dosage (2.5 mg/L + 24 h demand) for NDMA formation test was determined by calculating the respective chloramine dosage for the 24 h total chlorine residual of 2.5 mg/L using the linear regression line equations (details are presented in Appendix 1). Thus, the chloramine dosage for the NDMA formation tests in Lake Ontario water and Otonabee River water were determined to be 2.8 and 3.0 mg/L, respectively.

3.3.2 NDMA Formation Test

1 L amber bottles were used to perform the NDMA formation tests at room temperature (22 °C). The bottles were first filled with the selected water type to ¾ full; cation stock solutions were then added to achieve the designated dosage. After a half-hour interval to ensure good mixing and equilibration, 50 µL of ranitidine stock solution (0.1M) was dosed to achieve a target concentration of 5 nM (1572 ng/L). Afterwards, a calculated volume of freshly made monochloramine working solution was dosed to achieve the target concentrations of 2.8 mg/L for Lake Ontario water and 3.0 mg/L for Otonabee River water. The bottles were filled to be head-space free with the selected water and incubated for the designated contact times. NDMA
formation in the bottle was terminated by adding L-ascorbic acid powder (approximately 200 mg/L sample). The preparation of blank control samples using the respective matrix was similar to the above, only without the addition of ranitidine. Net NDMA formation from ranitidine was obtained by subtracting the average NDMA concentration in the blank control samples from the average NDMA concentration in the triplicate samples.

3.4 QA/QC Protocol

For each batch of experiments, a 6-level calibration curve was prepared with the selected water to calculate the NDMA concentration in the chloraminated samples (details are presented in Appendix 2). 1 L amber bottles were filled with 500 mL of the selected water to prepare samples for the calibration curve. 50 ng/L of $d_6$-NDMA was dosed to each sample as the internal standard. Except for the blank sample (i.e., 0 ng/L NDMA), NDMA stock solutions (1 or 10 mg/L) were dosed to achieve the five target concentrations: 50, 100, 200, 300, and 500 ng/L. The 1 mg/L NDMA stock solution was used to prepare the low concentrations (≤ 200 ng/L), while the other two concentrations were prepared using the 10 mg/L stock solution. In the NDMA formation tests, samples were prepared in triplicate and blank control samples (without addition of ranitidine) were prepared in either duplicate or triplicate.

One way Analysis of Variance (ANOVA), using cation concentration as the single factor and NDMA formation kinetics as the response, was applied to determine the correlation between cation concentration and NDMA formation. Depending on the one way ANOVA test results, Tukey's HSD Post Hoc Test was performed to indicate the difference in NDMA formation from different pairs of two cation concentration conditions.
4 Cation Impact on NDMA Formation from Ranitidine in Milli-Q® Water

4.1 Experimental Design

In order to study the impact of cation (i.e., Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^{+}\)) on N-Nitrosodimethylamine (NDMA) formation from ranitidine in real water matrices, the interactions between cations and ranitidine upon chloramination must first be investigated. Therefore, Milli-Q® water with the addition of cations at various dosages (up to 200 mg/L of Ca\(^{2+}\), 100 mg/L of Mg\(^{2+}\), and 50 mg/L of Na\(^{+}\)) was used to perform these initial NDMA formation tests. In this chapter, the ranitidine dosage was selected at 5 nM (1.57 µg/L), which is high enough to form detectable levels of NDMA given the high conversion of ranitidine to NDMA. This concentration level of ranitidine also simulates the higher end of the real-world scenario, as a similar concentration was detected in a hospital effluent (Gómez et al., 2006).

NDMA formation potential (FP) tests were conducted in 1 L amber bottles without pH adjustment (the pH of Milli-Q water was measured as 5.7). For each cation dosage, blank control samples (without ranitidine) and triplicate test samples (with 5 nM of ranitidine) were prepared. The NDMA FP tests were performed with a monochloramine dosage of 2.5 mg/L. Following the desired reaction time, the residual chloramine was quenched by adding ascorbic acid (around 200 mg/L sample). Further details of the NDMA formation potential test are described in Section 3.3.2. The extraction, concentration, analysis, and quantification of the NDMA formed in the samples are described in Section 3.2.3. Net NDMA formation from ranitidine was obtained by subtracting the average NDMA concentration in the blank control samples from the average NDMA concentration in the triplicate samples.

4.1.1 Cation Addition Dosages

Since the three cations of interest almost always coexist in natural waters, in this experiment they were studied as a group in Milli-Q® water instead of as individuals. Given the initial cation concentration in Lake Ontario water and Otonabee River water, a similar concentration (X: 40 mg/L of Ca\(^{2+}\), 20 mg/L of Mg\(^{2+}\), and 10 mg/L of Na\(^{+}\)) was tested in Milli-Q® water as well as several higher levels (2X, 5X). The dosages are summarized in Table 4-1, and the respective equivalent hardness was calculated based on the concentration of Ca\(^{2+}\) and Mg\(^{2+}\).
<table>
<thead>
<tr>
<th>Cation Addition Dosages Applied to Milli-Q® Water Samples</th>
<th>0X</th>
<th>X</th>
<th>2X</th>
<th>5X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca$^{2+}$ (mg/L)</td>
<td>0</td>
<td>40</td>
<td>80</td>
<td>200</td>
</tr>
<tr>
<td>Mg$^{2+}$ (mg/L)</td>
<td>0</td>
<td>20</td>
<td>40</td>
<td>100</td>
</tr>
<tr>
<td>Na$^+$ (mg/L)</td>
<td>0</td>
<td>10</td>
<td>20</td>
<td>50</td>
</tr>
<tr>
<td>Equivalent Hardness (mg/L as CaCO$_3$)</td>
<td>0</td>
<td>183</td>
<td>366</td>
<td>915</td>
</tr>
</tbody>
</table>

$X = 40$ mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations.

For each dosage, triplicate samples (with ranitidine) and triplicate blank control samples (without ranitidine) were prepared.

### 4.1.2 Contact Time Selection

Shen and Andrews (2011b) have examined the NDMA formation from ranitidine in Milli-Q® water with the same ranitidine (5 nM) and monochloramine (2.5 mg/L) dosages. The experiment was conducted with contact times up to 24 h. By fitting the observed NDMA formation into a modified dose-response model, the contact time required to reach a 50% of the ultimate NDMA formation ($t_{50\%}$) was determined as approximately 6 h. Also, NDMA has been found to be rapidly formed in the period of 2 h to 8 h. If there was to be any impact from cations on NDMA formation, it was expected to be most easily identified within the rapid growth phase. Allowing for a possible shift (e.g., ±2 h) of the $t_{50\%}$ and ensuring the selected contact time was within the rapid growth phase, instead of 6 h, the contact time in this experiment was selected to be 4 h.

### 4.2 Results and Discussion

#### 4.2.1 NDMA in the Blank Control Samples

NDMA formation following 4 h chloramination at different cation addition dosages in the blank control samples is shown in Figure 4-1 (raw data are presented in Appendix 3); the error bars represent the standard deviation of the variability in the triplicates. The NDMA concentration was found to be the same (around 25 ng/L) under the different cation concentrations ($p$-value = 0.520; one way ANOVA, 95% confidence level). In the blank control samples, only monochloramine and different levels of cations were dosed. Since a NDMA precursor must contain a nitrogen atom in its molecule, cations are not likely to form NDMA upon
chloramination. Therefore, NDMA detected in such samples must have been generated from the potential NDMA precursors in the Milli-Q® water or was already present in Milli-Q® water.

Figure 4-1. NDMA formation upon 4 h chloramination under different cation addition dosages in the blank control samples (MQ, preformed monochloramine = 2.5 mg/L; error bars presented with standard deviation (n = 3))

Compared with natural water sources, which usually contain total organic carbon (TOC) at mg/L levels, the TOC level in Milli-Q® was much lower (< 5 µg/L (EMD Millipore, 2013)). Also, given that NDMA concentration detected in the drinking water systems are generally below 10 ng/L (Russell et al., 2012), it is unlikely that the TOC in the Milli-Q® water could result in an NDMA formation of approximately 25 ng/L. Such concentration was more likely related to the NDMA originally occurring in Milli-Q®. The ion exchange resin in the Milli-Q® reactor was likely the source of NDMA: Najm and Trussell (2001) found that up to 130 ng/L of NDMA can be formed via the contact between various ion exchange resins and deionized water. Even though the ultraviolet (UV) light equipped in the Milli-Q® reactor was capable of removing NDMA, its removal efficiency deteriorated as it aged. Although the NDMA detected in the
Milli-Q® blank tests was higher than expected or desired, it was relatively constant and was lower than the levels formed when ranitidine was present.

4.2.2 NDMA Formation from Ranitidine

NDMA formation from ranitidine following 4 h chloramination at different cation addition dosages in Milli-Q® water is shown in Figure 4-2 (raw data is presented in Appendix 3); the error bars represent the standard deviation of the variability in the triplicates. The NDMA molar conversion was calculated using the equation below; 74 g/mol is the molar mass of NDMA, while 5 nM was the ranitidine dosage that was employed.

\[
\text{NDMA molar conversion (\%) = } \frac{\text{NDMA concentration (ng/L)}}{74 \text{ g/mol} \cdot 5 \text{ nM}} \times 100 \% \quad \text{(Eq. 4-1)}
\]

Figure 4-2. NDMA formation from ranitidine upon 4 h chloramination under different cation addition dosages in Milli-Q® water (5 nM ranitidine, preformed monochloramine = 2.5 mg/L; error bars were presented with standard deviation (n = 3))

The addition of excess cations into Milli-Q® water did not affect the NDMA formation from ranitidine upon 4 h chloramination (p-value = 0.345; one way ANOVA, 95 % confidence level).
Since the reaction of ranitidine to form NDMA has been found to be very rapid (approximately 4 h; Shen and Andrews, 2011b), if there was any significant impact from the cations on the NDMA formation, it should have been identified at 4 h. Therefore, the results obtained at 4 h contact indicate that cation concentration does not affect the NDMA formation in Milli-Q® water.

### 4.3 Summary

In this chapter, NDMA formation from ranitidine following 4 h chloramination was conducted in Milli-Q® water with various additions of cations (i.e., 0X, X, 2X, and 5X). Little difference was observed in the NDMA molar conversions under various cation concentrations, suggesting that the interactions between cations and ranitidine, if any, has a negligible effect on NDMA formation from ranitidine.
5 Cation Impact on NDMA Formation from Ranitidine in Lake Ontario Water and Otonabee River Water

The results in Chapter 4 have suggested that cations do not affect the conversion of ranitidine to N-Nitrosodimethylamine (NDMA) in Milli-Q® water, but the question remains whether it is still true when it comes to natural waters. Previous literature has indicated that the possible interactions between natural organic matter (NOM) and ranitidine could affect the formation of NDMA upon chloramination (Selbes et al., 2013; Shen and Andrews, 2011b). In addition, cations have been found to affect the aggregation and dissolution processes of NOM (Brigante et al., 2009; Kloster et al., 2013). In this chapter, two types of natural water sources, lake and river water, were selected to investigate the cation impact on NDMA formation from ranitidine: Lake Ontario water has a low total organic carbon (TOC) level whereas Otonabee River water has a higher TOC level.

5.1 Experimental Details

Previous literature has suggested that the presence of NOM could slow down the formation of NDMA due to the possible interactions between NOM and ranitidine (Shen and Andrews, 2011b). Theoretically, if cations could interfere in such interactions, they could affect the NDMA formation kinetics. In addition, even though it has been suggested that natural water matrices do not affect the ultimate yield of NDMA (Shen and Andrews, 2011b), it remains unknown whether higher cation concentrations than those experienced in previous research could lead to a different outcome.

Therefore, experiments were conducted to investigate the impact of cations on the kinetics and ultimate yield of NDMA formation in lake and river water matrices. NDMA formation from ranitidine at various cation concentrations (up to 202 mg/L of Ca²⁺, 90.1 mg/L of Mg²⁺, and 50.4 mg/L of Na⁺) in Lake Ontario water and Otonabee River water was monitored for up to 96 h using formation potential (FP) tests that were conducted in 1 L amber bottles without pH adjustment. Similar to that for experiments using Milli-Q® water, ranitidine dosage for lake and river water experiments was selected as 5 nM (1.57 µg/L). For each combination of water matrix and cation dosage, blank control samples (without ranitidine) and triplicate test samples (with ranitidine) were prepared. The NDMA FP tests were performed with a monochloramine
dosage of 2.8 mg/L for lake water and 3.0 mg/L for river water. Following each desired reaction time, the residual chloramine was quenched by adding ascorbic acid (approximately 200 mg/L sample). Further details concerning the NDMA formation potential test are described in Section 3.3.2. The extraction, concentration, analysis, and quantification of the NDMA formed in the samples are introduced in Section 3.2.3. Net NDMA formation from ranitidine was obtained by subtracting the average NDMA concentration in the blank control samples from the average NDMA concentration in the triplicate samples.

5.1.1 Raw Water Quality

Raw, untreated water was collected from two water utilities in Ontario: Lake Ontario water from the lab at the Lorne Park Water Treatment Plant (Mississauga, Ontario) on July 13th and September 4th, and Otonabee River water from the intake line at the Peterborough Water Treatment Plant on Oct 28th. Water quality parameters for both waters were measured in duplicate and the average values are summarized in Table 5-1. The two waters exhibited different DOC levels, but similar portions of different NOM fractions were found: the DOC level in Lake Ontario water was nearly 1/3 of that in the river water; the portions of the five NOM fractions (biopolymers, humic substances, building blocks, low molecular weight (LMW) acids, and LMW neutrals) of the DOC were found to be similar in the two waters. For example, the humic substances found in the lake and river water were both approximately 50 % of the corresponding DOC (2.1 ± 0.2 and 5.9 ± 0.1 mg/L, respectively).
Table 5-1. Water quality of Lake Ontario water and Otonabee River water

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Lake Ontario water</th>
<th>Otonabee River water</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.9 ± 0.1</td>
<td>8.0 ± 0.1</td>
</tr>
<tr>
<td>UV$_{254}$</td>
<td>0.028 ± 0.001</td>
<td>0.137 ± 0</td>
</tr>
<tr>
<td>Alkalinity (mg/L)</td>
<td>94.0 ± 0</td>
<td>76.5 ± 0.7</td>
</tr>
<tr>
<td>TOC$^a$ (mg/L)</td>
<td>2.0 ± 0.1</td>
<td>5.4 ± 0.1</td>
</tr>
<tr>
<td>DOC$^b$ (mg/L)</td>
<td>2.1 ± 0.2</td>
<td>5.9 ± 0.1</td>
</tr>
<tr>
<td>Hydrophobic DOC (mg/L)</td>
<td>0.29 ± 0.12</td>
<td>0.48 ± 0.24</td>
</tr>
<tr>
<td>Biopolymers (mg/L)</td>
<td>0.28 ± 0.01</td>
<td>0.54 ± 0.03</td>
</tr>
<tr>
<td>Humic substances (mg/L)</td>
<td>0.97 ± 0.07</td>
<td>3.3 ± 0.06</td>
</tr>
<tr>
<td>Building blocks (mg/L)</td>
<td>0.31 ± 0.12</td>
<td>0.85 ± 0.09</td>
</tr>
<tr>
<td>LMW acids (mg/L)</td>
<td>0.20 ± 0</td>
<td>0.54 ± 0.17</td>
</tr>
<tr>
<td>LMW neutrals (mg/L)</td>
<td>0.06 ± 0.01</td>
<td>0.16 ± 0</td>
</tr>
</tbody>
</table>

a. Aurora 1030 TOC analyzer (O.I. Analytical, College Station, Texas)
b. Liquid Chromatography-Organic Carbon Detection (LC-OCD)

5.1.2 Cation Addition Dosages

Cation (i.e., Ca$^{2+}$, Mg$^{2+}$, and Na$^+$) concentrations in surface water sources usually vary widely. The Na$^+$ level in drinking water has been suggested to be typically less than 20 mg/L (WHO, 2003). The Ca$^{2+}$ and Mg$^{2+}$ levels in surface water sources in different parts of the world have been reported to have mean values of 34 ± 21 and 10 ± 8 mg/L, respectively (WHO, 2009), while higher levels of those ions have been found in ground water sources (52 ± 24 and 20 ± 13 mg/L, respectively). Given that surface and ground water are the major water sources for drinking water, in this study, a combination of 40 mg/L of Ca$^{2+}$, 20 mg/L of Mg$^{2+}$, and 10 mg/L of Na$^+$ was used as an approximation of the average cation concentrations of the two water types and represented as “X” to help set up the experimental design parameters. Also, the cation concentrations of X were close to those found originally in the lake (42.3 mg/L of Ca$^{2+}$, 10.1 mg/L of Mg$^{2+}$, and 10.4 mg/L of Na$^+$) (Sadmani et al., 2014) and river (39.3 mg/L of Ca$^{2+}$, 3.1 mg/L of Mg$^{2+}$, and 4.7 mg/L of Na$^+$) (Sadmani, 2013) water. In the current experiments, up to 4X cations were added to the raw water samples, the result being that the total cation concentrations in both waters was up to approximately 5X (the highest addition dosage applied...
in Milli-Q® water; see Chapter 4). In addition to the dosages of X and 4X, several intermediate cation dosages were also employed. The same cation addition dosages were applied to both waters, as listed in Table 5-2; the final cation concentrations in the lake and river water samples that were used in experiments are given in Table 5-3 and Table 5-4, respectively.

### Table 5-2. Cation addition dosages applied to Lake Ontario water and Otonabee River water samples

<table>
<thead>
<tr>
<th></th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca$^{2+}$ (mg/L)</td>
<td>0</td>
<td>10</td>
<td>20</td>
<td>40</td>
<td>80</td>
<td>160</td>
</tr>
<tr>
<td>Mg$^{2+}$ (mg/L)</td>
<td>0</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>40</td>
<td>80</td>
</tr>
<tr>
<td>Na$^+$ (mg/L)</td>
<td>0</td>
<td>2.5</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>40</td>
</tr>
<tr>
<td>Equivalent Hardness (mg/L as CaCO$_3$)</td>
<td>0</td>
<td>46.3</td>
<td>92.5</td>
<td>183</td>
<td>366</td>
<td>732</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations

### Table 5-3. Total cation concentrations in Lake Ontario water samples following different cation additions

<table>
<thead>
<tr>
<th></th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca$^{2+}$ (mg/L)</td>
<td>42.3</td>
<td>52.3</td>
<td>62.3</td>
<td>82.3</td>
<td>122</td>
<td>202</td>
</tr>
<tr>
<td>Mg$^{2+}$ (mg/L)</td>
<td>10.1</td>
<td>15.1</td>
<td>20.1</td>
<td>30.1</td>
<td>50.1</td>
<td>90.1</td>
</tr>
<tr>
<td>Na$^+$ (mg/L)</td>
<td>10.4</td>
<td>12.9</td>
<td>15.4</td>
<td>20.4</td>
<td>30.4</td>
<td>50.4</td>
</tr>
<tr>
<td>Equivalent Hardness (mg/L as CaCO$_3$)</td>
<td>148</td>
<td>194</td>
<td>240</td>
<td>331</td>
<td>514</td>
<td>880</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations
Table 5-4. Total cation concentrations in Otonabee River water samples following different cation additions

<table>
<thead>
<tr>
<th></th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca(^{2+}) (mg/L)</td>
<td>39.3</td>
<td>49.3</td>
<td>59.3</td>
<td>79.3</td>
<td>119</td>
<td>199</td>
</tr>
<tr>
<td>Mg(^{2+}) (mg/L)</td>
<td>3.1</td>
<td>8.1</td>
<td>13.1</td>
<td>23.1</td>
<td>43.1</td>
<td>83.1</td>
</tr>
<tr>
<td>Na(^{+}) (mg/L)</td>
<td>4.7</td>
<td>7.2</td>
<td>9.7</td>
<td>14.7</td>
<td>24.7</td>
<td>44.7</td>
</tr>
<tr>
<td>Equivalent Hardness (mg/L as CaCO(_3))</td>
<td>111</td>
<td>157</td>
<td>203</td>
<td>295</td>
<td>478</td>
<td>845</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca\(^{2+}\), 20 mg/L Mg\(^{2+}\), and 10 mg/L Na\(^{+}\) of added cations

In the Guidelines for Canadian Drinking Water Quality Hardness (1979), water with a hardness level within the range of 120 to 180 mg/L as CaCO\(_3\) would be categorized as “hard” water; roughly, the lake and river water with 0X and 0.25X additions could be considered as such type. In addition, water with a hardness level over 200 mg/L as CaCO\(_3\) has been categorized as “very hard”, and if the hardness level is higher than 500 mg/L, water would normally be considered unacceptable for domestic purposes (MOE, 2007). Accordingly, the lake and river water with 0.5X, X, and 2X additions could be categorized as “very hard”. The hardness level in both waters with 4X cation addition have been so rarely seen in surface water that they could be considered as extreme scenarios.

5.1.3 Model Used for NDMA Formation Kinetics Analysis

The reaction kinetics of NDMA formation from amine-based pharmaceuticals has been suggested to follow a pattern which is comprised of an initial lag period, a subsequent rapid increase phase, and a final plateau phase; such pattern could be described using a model proposed by Shen and Andrews (2011b) as given below:

\[
Y = \begin{cases} 
0 & (t = 0) \\
\frac{\theta}{1 + 10^k(Lag-t)} & (t > 0)
\end{cases}
\]

where Y is the NDMA molar conversion at the selected contact time (t); \(\theta\) is the ultimate NDMA molar conversion which equals to \(Y(t \rightarrow +\infty)\) and observed as the plateau value of an NDMA kinetic test; \(k\) is the pseudo-first order reaction rate constant (h\(^{-1}\)); Lag (h) is the time required to achieve 50% of the ultimate NDMA molar conversion. The NDMA molar conversion has been
arbitrarily set to 0 at t = 0 to represent the starting point of the reaction. Although the proposed model does not always go through the point of (0, 0) unless sufficient data can be obtained during the short period right after the reaction was triggered, the model has been found to fit the experimental data well with correlation coefficients (R²) higher than 0.95 for all tested pharmaceuticals including ranitidine (Shen and Andrews, 2011b). In this chapter, the model was applied to describe the reaction kinetics of ranitidine to form NDMA. For each combination of water matrix and cation level, the model was built based on at least six experimental data points. GraphPad Prism 5® software was applied to fit the NDMA formation curves as well as to estimate the respective model parameters.

5.2 Results with Lake Ontario Water

5.2.1 General Experimental Results

NDMA formation from ranitidine (5 nM) in Lake Ontario water at each cation level was monitored after 4, 8, 12, 16, 24, 48, and 72 h of reaction time. Samples with 0X, X, and 4X cation additions were also monitored at 2 h to help improve later modeling efforts for the early stages of reaction. The molar conversion of NDMA from ranitidine was calculated using Equation 4-1 and results are summarized in Table 5-5. Raw data are presented in Appendix 3.
Table 5-5. NDMA molar conversion from ranitidine in Lake Ontario water at different contact times under various cation addition dosages

<table>
<thead>
<tr>
<th>Contact time (h)</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>1.4% (0.1%)</td>
<td>N/A</td>
<td>N/A</td>
<td>1.0% (0.1%)</td>
<td>N/A</td>
<td>1.0% (0.1%)</td>
</tr>
<tr>
<td>4</td>
<td>11.5% (0.7%)</td>
<td>0.7% (0.6%)</td>
<td>0% (0.6%)</td>
<td>4.5% (1.0%)</td>
<td>2.7% (2.1%)</td>
<td>7.2% (1.6%)</td>
</tr>
<tr>
<td>8</td>
<td>40.5% (1.2%)</td>
<td>20.8% (1.8%)</td>
<td>12.4% (1.4%)</td>
<td>26.0% (1.8%)</td>
<td>12.3% (2.6%)</td>
<td>34.4% (2.7%)</td>
</tr>
<tr>
<td>12</td>
<td>68.8% (4.4%)</td>
<td>74.0% (12.0%)</td>
<td>74.7% (5.4%)</td>
<td>37.9% (2.1%)</td>
<td>65.7% (9.7%)</td>
<td>47.3% (4.0%)</td>
</tr>
<tr>
<td>16</td>
<td>77.7% (9.2%)</td>
<td>73.8% (2.9%)</td>
<td>47.7% (3.2%)</td>
<td>45.4% (5.3%)</td>
<td>51.2% (5.1%)</td>
<td>57.2% (2.2%)</td>
</tr>
<tr>
<td>24</td>
<td>79.5% (2.2%)</td>
<td>86.2% (2.9%)</td>
<td>54.3% (11.3%)</td>
<td>53.5% (1.8%)</td>
<td>61.6% (10.3%)</td>
<td>69.3% (5.9%)</td>
</tr>
<tr>
<td>48</td>
<td>84.7% (3.0%)</td>
<td>82.5% (6.6%)</td>
<td>83.7% (3.8%)</td>
<td>71.9% (0.5%)</td>
<td>80.1% (10.0%)</td>
<td>77.4% (2.8%)</td>
</tr>
<tr>
<td>72</td>
<td>85.4% (3.5%)</td>
<td>70.5% (0.9%)</td>
<td>78.8% (2.5%)</td>
<td>84.5% (1.5%)</td>
<td>76.2% (0.9%)</td>
<td>85.9% (2.8%)</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca\(^{2+}\), 20 mg/L Mg\(^{2+}\), and 10 mg/L Na\(^+\) of added cations
* Numbers in the brackets represent the standard deviation from triplicate test

At the beginning of the test (0 to 4 h), the formation of NDMA was so slow that the NDMA conversions were found to be generally below 10%. The accumulation of NDMA in the lake water became rapid from 8 h and started to slow down at 24 h, so the period from 8 to 24 h might be referred to as the rapid growth phase of ranitidine to form NDMA. For each cation level, similar NDMA molar conversions were found in the 48 h and 72 h samples, suggesting the plateau of NDMA formation had been reached in all cases. As suggested by Shen and Andrews (2011b), these experimental data (covering the initial lag period, the rapid growth phase, and the ultimate plateau phase) could be used to acquire reliable estimates for the three kinetic model parameters Lag, k, and \(\theta\), respectively.

The NDMA molar conversions with their respective standard deviations were input to the software and fitted to the model. As an example, the NDMA molar conversions over time for ranitidine in Lake Ontario water without cation addition are given in Figure 5-1, where the markers are the measured data, and the curve is the estimated result generated from the model. The curve was well-fitted to the experimental data, with the correlation coefficient \((R^2)\) higher.
than 0.95. Further discussion about the measured data and the model results are provided in the following sections of this thesis.

![Graph](image)

**Figure 5-1.** NDMA molar conversion in Lake Ontario water without cation addition (5 nM ranitidine, preformed monochloramine = 2.8 mg/L; error bars present one standard deviation (n = 3))

In some cases where there was sufficient reason to do so, potential outliers were excluded. For example, NDMA conversions in the 12 h samples with 0.25X, 0.5X, and 2X cation additions were so high that they were not used for model analysis. The possible cause for such odd results was believed to be the change of chloramine composition in the monochloramine working solution. Due to practical feasibility, the reactions of the 12 h samples (of 0.25X, 0.5X, and 2X) were started at night, while those of the other samples were started much earlier that day in the morning; the disproportionation of monochloramine in the working solution (prepared in the morning) likely caused the presence of dichloramine which has been found to significantly enhance NDMA formation (Mitch et al., 2005).
5.2.2 Cation Impact on Model Parameters

The kinetic model parameters together with their respective standard deviations were generated by the GraphPad Prism 5® software and are summarized in Table 5-6. The values of $R^2$ in the table are all higher than 0.9, indicating that the modeled results were well-fitted to the measured data.

<table>
<thead>
<tr>
<th>Model Parameter</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\theta$</td>
<td>82.5 %</td>
<td>79.3 %</td>
<td>80.0 %</td>
<td>76.4 %</td>
<td>75.5 %</td>
<td>78.1 %</td>
</tr>
<tr>
<td>(1.3 %)*</td>
<td>(2.0 %)</td>
<td>(3.5 %)</td>
<td>(3.1 %)</td>
<td>(2.8 %)</td>
<td>(2.5 %)</td>
<td></td>
</tr>
<tr>
<td>Lag (h)</td>
<td>8.2 (0.2)</td>
<td>10.1 (0.7)</td>
<td>16.6 (1.2)</td>
<td>14.4 (1.1)</td>
<td>14.0 (0.9)</td>
<td>10.8 (0.6)</td>
</tr>
<tr>
<td>$k$ (h$^{-1}$)</td>
<td>0.20 (0.02)</td>
<td>0.22 (0.05)</td>
<td>0.08 (0.01)</td>
<td>0.07 (0.01)</td>
<td>0.11 (0.02)</td>
<td>0.11 (0.01)</td>
</tr>
<tr>
<td>$R^2$</td>
<td>0.985</td>
<td>0.972</td>
<td>0.938</td>
<td>0.937</td>
<td>0.951</td>
<td>0.954</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations

* Numbers in these brackets represent the standard deviation of each parameter

5.2.2.1 Cation Impact on Ultimate Molar Conversion ($\theta$)

The estimated ultimate NDMA molar conversion ($\theta$) values obtained at each cation level are summarized in Figure 5-2. Even though not all of the error bars overlapped, one-way analysis of variance (ANOVA) tests (provided in Appendix 4) suggested that they were not significantly different from each other (p-value = 0.065; one way ANOVA, 95 % confidence level). Tukey's HSD post hoc test results (provided in Appendix 4) also confirmed that there was no significant difference in $\theta$ for each pair of cation levels. These results suggested that cations did not affect the ultimate NDMA formation from ranitidine in Lake Ontario water, which is consistent with results that have been reported by Shen (2013).
Figure 5-2. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in Lake Ontario water with various cation additions (0X = 148 mg/L as CaCO$_3$ hardness; error bars represent one standard deviation (n=3))

5.2.2.2 Cation Impact on the Lag Parameter

The estimated Lag times for NDMA formation at each cation level are summarized in Figure 5-3. The cation level was found to significantly affect the Lag values (p-value < 0.0001; one way ANOVA, 95 % confidence level). As the cation concentration increased, the initial lag period first increased and subsequently decreased. The highest Lag value (16.6 h) was obtained after a 0.5X cation addition (an increase in hardness from 148 to 240 mg/L as CaCO$_3$). Since the Lag value represents the time required to achieve 50 % of the ultimate NDMA formation ($t_{50\%}$) and since a higher Lag value would indicate a longer initial inhibition period in the reaction kinetics of NDMA formation, the current results indicate that the initial reaction between ranitidine and chloramine in Lake Ontario water was inhibited the most upon 0.5X cation addition.
Figure 5-3. Estimation of model parameter Lag for NDMA formation from ranitidine in Lake Ontario water with various cation additions (0X = 148 mg/L as CaCO\textsubscript{3} hardness; error bars represent one standard deviation (n=3))

Tukey's HSD test was also applied to analyze the Lag values in pairs. Most of these pairwise comparisons of Lag data for the additions of extra cations in lake water were found to suggest significant differences in the Lag values (p-value < 0.05; Tukey's HSD test, 95 % confidence level) except for the addition of 0.25X (p-value > 0.05; Tukey's HSD test, 95 % confidence level). It was also noticed that the Lag values of some nearby cation levels were similar to each other. For example, the Lag values of samples with 0.5X and X cation addition were not considered significantly different (p-value > 0.05; Tukey's HSD test, 95 % confidence level). Further discussion of the pair test results will be given in Section 5.2.3.

5.2.2.3 Cation Impact on Reaction Rate Constant (k)

The model parameter k indicates the reaction rate constant for NDMA formation in the rapidly increasing phase and its value at each cation level are summarized in Figure 5-4. The addition of excess cations (≥ 0.5X or 240 mg/L as CaCO\textsubscript{3} in hardness) was found to significantly affect the parameter k (p-value < 0.0001; one-way ANOVA, 95 % confidence level). Regardless of the
large variance in the k values for samples with 0.25X cation addition, as the cation concentration increased, the k value first largely decreased to a minimum at approximately 0.5X to X and was followed by a slight increase in k upon further increases in cations. Although the difference in the k values between every two cation addition dosages of the 0.5X, X, 2X, and 4X was insignificant (p-value > 0.05; Tukey’s HSD test, 95 % confidence level), the k value was so sensitive to the initial addition of cations that it dropped by nearly 60 % with the addition of only 0.5X.

![Figure 5-4. Estimation of model parameter k for NDMA formation from ranitidine in Lake Ontario water with various cation additions (0X = 148 mg/L as CaCO$_3$ hardness; error bars represent one standard deviation (n=3))]()

5.2.3 Evaluation of Cation Impact on NDMA Formation Kinetics under Moderate and Extremely High Hardness Conditions

The model that quantitatively describes the formation of NDMA from ranitidine is a function of three parameters: θ, k, and Lag. Since the discussion above has suggested that cations affect the kinetic model parameters in different ways, various cation concentrations would be expected to
result in different NDMA formation kinetics details which, except for the ultimate molar conversions ($\theta$), is what was observed.

It was further observed that the effects of cations could be divided into two general categories: effects at “moderate or high” hardness and effect at “extremely high” hardness. To illustrate these two general sets of effects, the NDMA molar conversions over time for ranitidine under no addition, X addition, and 4X addition of cations are summarized in Figure 5-5. The markers in the figure represent the measured NDMA molar conversions, while the curves are the estimated trends generated from the model (the same format was also applied for Figure 5-6 and Figure 5-7). Lake Ontario water itself was considered as “hard” water, and with X addition of cations it was considered “very hard”. It could be seen from Figure 5-5 that the addition of X cations prolonged the initial lag period of the reaction forming NDMA, yet the initial lag period was shortened with further additions of cations. The initial lag period of samples without cation addition was the shortest. These observations seemed to indicate that, in the first couple of hours after the chloramination started, the risk of NDMA formation would be larger in lake water itself than that in lake water with higher cation concentrations.
After the initial lag period, the reaction of forming NDMA started to become rapid. Without the addition of cations, the NDMA formation from ranitidine in Lake Ontario water reached its plateau within 24 h, while the addition of cations generally postponed the maximum NDMA formation. For samples with addition of X cations, the time to reach the ultimate NDMA formation was nearly twice as that for samples with no cation addition. In fact, the time required to reach the ultimate NDMA formation was mostly related to the reaction rate within the rapid growing phase. In the post-hoc analysis, significant difference was found in the pseudo-first order reaction rate constant for samples with and without cation addition (p-value < 0.05; Tukey's HSD test, 95 % confidence level); yet the difference between the reaction rate constants for samples with cation additions (X and 4X) was found insignificant (p-value > 0.05; Tukey's HSD test, 95 % confidence level). Addition of even small amounts of additional cations reduced the overall reaction rate.
The one-way ANOVA and Tukey’s HSD test results (details are given in Table A4-2 in Appendix 4) together with the shapes of the three curves seemed to suggest that the addition of cations affects NDMA formation kinetics differently within two general cation addition dosage ranges: 0X to X and X to 4X. Accordingly, the cation impact on NDMA formation kinetics will be discussed with regards to these two ranges, respectively, in the following sections.

Varying the cation addition dosages from ambient levels (0X) to double their ambient levels (X) was found to affect the NDMA formation kinetics in a different pattern. The NDMA molar conversions in lake water with cation additions of 0X to X are given in Figure 5-6. As shown in the figure, the addition of 0.25X cations did not affect the NDMA formation kinetics in terms of the model parameters ($\theta$, Lag, and k) (p-value > 0.05; Tukey's HSD test, 95 % confidence level). Similarly, the NDMA formation kinetics of samples with 0.5X and X cation additions were found to be comparable (p-values > 0.05; Tukey's HSD test, 95 % confidence level). Note that the hardness of lake water with 0.25X cation addition and lake water itself could be categorized as “hard”, while that of lake water with 0.5X and X cation addition would belong to the “very hard” category (Health Canada, 1979). Therefore, these results might suggest that in Lake Ontario water, the kinetics of NDMA formation from ranitidine was associated with the range of hardness to some extent.
Interestingly, the NDMA formation kinetics of samples with 0.25X and 0.5X additions were significantly different (p-values > 0.05 for Lag and \(k\); Tukey's HSD test, 95 % confidence level). The initial lag period of NDMA molar conversion under 0.5X cation addition was much longer than that under 0.25X addition; the time required for samples with 0.25X cation addition to reach the 50 % ultimate NDMA formation was only around half of that for samples with 0.5X addition. The significant difference in NDMA formation kinetics between these two conditions might suggest that ranitidine forming NDMA in Lake Ontario water was very sensitive to the addition of cations from 0.25X to 0.5X (194 to 240 mg/L as CaCO\(_3\) in hardness).

The NDMA molar conversions from ranitidine in Lake Ontario water with very high (X, 2X, and 4X) cation additions are given in Figure 5-7. As mentioned before, the abnormal high conversion at 12 h of the samples with 2X cation addition was considered as an outlier. Except for that data point, other measured data were close to the curves generated by the model.
Figure 5-7. NDMA molar conversions from ranitidine in Lake Ontario water with X, 2X, and 4X cation additions (5 nM ranitidine, preformed monochloramine = 2.8 mg/L; error bars were presented with standard deviation (n = 3))

The range of X to 4X was a transition of water hardness from “very hard” to unsuitable for domestic use (but included in this study to further examine the limits of the cation effects). In this range, the time required to reach the ultimate NDMA formation reduced as the cation concentration increased; in other words, increasing the cation addition dosages from X to 4X in lake water enhanced the initial NDMA formation from ranitidine. The scale of the reduction in the time required for ultimate formation was minimal between the additions of 2X and 4X, which can be reflected from their nearly parallel rapid growing phases. Tukey’s HSD test confirmed that the addition of cations in this range did not affect the pseudo-first reaction rate (k) of NDMA formation (p-value > 0.05; Tukey’s HSD test, 95 % confidence level). In another words, the rapid growth phase of NDMA formation was not sensitive to the changes in cation concentrations in the addition range of X to 4X.

The Lag value of the condition 4X was significantly different from that of the rest two conditions (p-value < 0.05; Tukey’s HSD test, 95 % confidence level). Since the three conditions tended to
have similar k values, the difference among the Lag value might be a result of the initial formation period (before the rapid growth phase): as shown in Figure 5-7, the initial lag period of 4X was shorter than that of the rest two conditions. However, such difference was not very obvious. In general, NDMA formation kinetics was not very sensitive to the increase of cation concentrations from X to 4X additions.

5.2.4 Summary

To summarize, in Lake Ontario water, the addition of cations did not affect the ultimate NDMA formation (θ), but did affect the NDMA formation kinetics (i.e., Lag and k). In general, the reaction rate of ranitidine to form NDMA first decreased and subsequently increased as the cation concentrations increased. The NDMA formation kinetics was very sensitive to the initial addition of cations, but less sensitive to the further increase of cation concentrations. Among the six selected addition dosages, the addition of 0.5X cations was found to result in the slowest formation of NDMA. In general, cation dosages from the same hardness category were found to have similar impact on the NDMA formation kinetics.

5.3 Results with Otonabee River Water

5.3.1 General Experimental Results

NDMA formation from ranitidine (5 nM) in Otonabee River water at each cation level was measured after 8, 16, 24, 48, 72, and 96 h. The molar conversion of NDMA from ranitidine was calculated using Equation 4-1 and results are summarized in Table 5-7 (raw data are presented in Appendix 3).
Table 5-7. NDMA molar conversion from ranitidine in Otonabee River water at different contact times under various cation addition dosages

<table>
<thead>
<tr>
<th>Contact time (h)</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>2.8 %</td>
<td>1.3 %</td>
<td>1.1 %</td>
<td>2.6 %</td>
<td>3.6 %</td>
<td>3.2 %</td>
</tr>
<tr>
<td></td>
<td>(1.0 %)*</td>
<td>(0.4 %)</td>
<td>(0.9 %)</td>
<td>(0.6 %)</td>
<td>(0.1 %)</td>
<td>(0.4 %)</td>
</tr>
<tr>
<td>16</td>
<td>4.2 %</td>
<td>3.8 %</td>
<td>3.2 %</td>
<td>4.5 %</td>
<td>5.1 %</td>
<td>5.0 %</td>
</tr>
<tr>
<td></td>
<td>(0.3 %)</td>
<td>(1.2 %)</td>
<td>(0.1 %)</td>
<td>(1.7 %)</td>
<td>(3.5 %)</td>
<td>(1.2 %)</td>
</tr>
<tr>
<td>24</td>
<td>11.5 %</td>
<td>10.4 %</td>
<td>6.3 %</td>
<td>9.5 %</td>
<td>11.4 %</td>
<td>13.3 %</td>
</tr>
<tr>
<td></td>
<td>(2.9 %)</td>
<td>(2.4 %)</td>
<td>(2.0 %)</td>
<td>(0.6 %)</td>
<td>(0.5 %)</td>
<td>(0.5 %)</td>
</tr>
<tr>
<td>48</td>
<td>20.9 %</td>
<td>19.9 %</td>
<td>20.0 %</td>
<td>20.8 %</td>
<td>23.8 %</td>
<td>34.3 %</td>
</tr>
<tr>
<td></td>
<td>(2.3 %)</td>
<td>(1.2 %)</td>
<td>(2.1 %)</td>
<td>(1.5 %)</td>
<td>(1.9 %)</td>
<td>(0.7 %)</td>
</tr>
<tr>
<td>72</td>
<td>58.3 %</td>
<td>56.5 %</td>
<td>48.2 %</td>
<td>50.9 %</td>
<td>53.7 %</td>
<td>65.5 %</td>
</tr>
<tr>
<td></td>
<td>(0.2 %)</td>
<td>(3.7 %)</td>
<td>(3.0 %)</td>
<td>(3.2 %)</td>
<td>(2.2 %)</td>
<td>(10.5 %)</td>
</tr>
<tr>
<td>96</td>
<td>65.5 %</td>
<td>64.8 %</td>
<td>68.9 %</td>
<td>67.2 %</td>
<td>66.4 %</td>
<td>72.7 %</td>
</tr>
<tr>
<td></td>
<td>(2.8 %)</td>
<td>(2.4 %)</td>
<td>(8.7 %)</td>
<td>(4.5 %)</td>
<td>(4.9 %)</td>
<td>(4.0 %)</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca\(^{2+}\), 20 mg/L Mg\(^{2+}\), and 10 mg/L Na\(^{+}\) of added cations

* Numbers in these brackets represent the standard deviation of each parameter

Comparing with that in Lake Ontario water, the formation of NDMA in Otonabee River water (0X) was much slower: less than 5 % formation was observed in the first 8 h. Consequently, the ultimate NDMA formation was not achieved until 96 h. The results were in accordance with the findings from Shen and Andrews (2011b): the slower formation kinetics was a result of the higher occurrence of NOM in the river water. All the measured data in the table were input to the GraphPad Prism 5\(^{\text{®}}\) software and fitted to the model to estimate the model parameters and generate NDMA formation kinetics curves. As an example, NDMA molar conversions over time for ranitidine in Otonabee River water without cation addition are summarized in Figure 5-8, where the markers are the measured data, and the curve is the estimated result generated from the model. The curve was well fitted to the experimental data, with the correlation coefficient (R\(^2\)) as 0.977. Other results of the model analysis are given and discussed in the following sections.
Figure 5-8. NDMA molar conversion in Otonabee River water without cation addition (5 nM ranitidine, preformed monochloramine = 3.0 mg/L; error bars represent one standard deviation (n = 3))

5.3.2 Cation Impact on Model Parameters

The estimated values of the three model parameters at various cation addition dosages are summarized in Table 5-8. As shown in the table, the correlation coefficient ($R^2$) for all cases were higher than 0.97, suggesting that the measured data were well-fitted to the curves generated by the model.
Table 5-8. Kinetic model parameter estimation for Otonabee River water samples

<table>
<thead>
<tr>
<th>Model Parameter</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>θ</td>
<td>70.5 %</td>
<td>69.2 %</td>
<td>78.2 %</td>
<td>75.9 %</td>
<td>72.9 %</td>
<td>76.6 %</td>
</tr>
<tr>
<td></td>
<td>(3.8 %)*</td>
<td>(3.1 %)</td>
<td>(5.1 %)</td>
<td>(3.8 %)</td>
<td>(3.4 %)</td>
<td>(3.5 %)</td>
</tr>
<tr>
<td>Lag (h)</td>
<td>55.6 (2.7)</td>
<td>56.2 (2.2)</td>
<td>64.6 (3.1)</td>
<td>61.5 (2.5)</td>
<td>57.1 (2.5)</td>
<td>49.7 (2.5)</td>
</tr>
<tr>
<td>k (h⁻¹)</td>
<td>0.03 (0.004)</td>
<td>0.03 (0.004)</td>
<td>0.03 (0.003)</td>
<td>0.03 (0.002)</td>
<td>0.03 (0.002)</td>
<td>0.03 (0.004)</td>
</tr>
<tr>
<td>R²</td>
<td>0.977</td>
<td>0.982</td>
<td>0.984</td>
<td>0.989</td>
<td>0.987</td>
<td>0.977</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca²⁺, 20 mg/L Mg²⁺, and 10 mg/L Na⁺ of added cations
* Numbers in these brackets represent the standard deviation of each parameter

5.3.2.1 Cation Impact on Ultimate Molar Conversion (θ)

The estimated values of NDMA molar conversion (θ) at the selected cation addition dosages are summarized in Figure 5-9. The error bars represent the standard deviation of the triplicate analysis. The cation concentrations were found to have insignificant impact on the ultimate NDMA formation from ranitidine in Otonabee River water matrices (p-value = 0.082; one-way ANOVA, 95 % confidence level). Moreover, Tukey’s HSD test results (given in Appendix 4) have confirmed that there was no significant difference in θ between each pair of cation levels (p-value > 0.05; Tukey’s HSD test, 95 % confidence level). These results suggested that cations did not affect the ultimate NDMA formation from ranitidine in Otonabee River water.
5.3.2.2 Cation Impact on the Lag Parameter

The estimated Lag times for NDMA formation at various cation addition dosages are summarized in Figure 5-10. The addition of cations has been found to significantly affect the Lag values (p-value = 0.0003; one-way ANOVA test, 95% confidence level). The Lag values first increased, peaked at 0.5X cation addition (an increase in hardness from 111 to 203 mg/L as CaCO$_3$), and gradually decreased as the cation concentrations increased. An initial 0.25X cation addition in river water did not result in significant change of the Lag value (p-value > 0.05; Tukey's HSD test, 95% confidence level); while a further 0.25X cation addition (to reach 0.5X addition) has raised the Lag value by nearly 9 h and reached the peak value. After that, as the cation concentration increased, the Lag values decreased, but the difference in the Lag values between every two nearby cation dosages was found insignificant (p-value > 0.05; Tukey's HSD test, 95% confidence level) except for that between 2X and 4X addition. It might suggest that...
the Lag value was not sensitive to the increase of cation concentration from 0.5X addition to above.

![Figure 5-10. Estimation of model parameter Lag for NDMA formation from ranitidine in Otonabee River water with various cation additions (0X = 111 mg/L as CaCO₃ hardness; error bars represent one standard deviation (n=3))](image)

5.3.2.3 Cation Impact on Reaction Rate Constant (k)

The estimated value of reaction rate constant (k) at each cation addition dosage is summarized in Figure 5-11. Apparently, most of the error bars were overlapped. Statistical analysis results (details are presented in Appendix 4) have suggested that cation concentration did not affect the pseudo-first order reaction rate constant of NDMA formation from ranitidine in Otonabee River water (p-value = 0.051; one-way ANOVA, 95% confidence level). Moreover, Tukey’s HSD test results (provided in Appendix 4) have confirmed that there was no significant difference between each pair of the obtained k values (p-values > 0.05; Tukey’s HSD test, 95% confidence level). However, the little variance observed in the k values of river water samples might due to the fact that not enough data points were captured during the rapid growth phase of the NDMA formation. From Table 5-7, under each cation dosage, the rapid growing phase was roughly
from 24 h to 72 h, and the most rapid formation of NDMA was observed within the 24 h interval between contact times of 48 h and 72 h. Therefore, it is possible that the difference in the k values was not well revealed.

![Graph](image)

**Figure 5-11.** Estimation of model parameter k for NDMA formation from ranitidine in Otonabee River water with various cation additions (0X = 111 mg/L as CaCO$_3$ hardness; error bars represent one standard deviation (n=3))

5.3.3 Evaluation of Cation Impact on NDMA Formation Kinetics under Moderate and Extremely High Hardness Conditions

In Otonabee River water, the cation impact was only found on the model parameter Lag with a 95 % confidence level. Since the kinetics of NDMA formation is a function of the three parameters, it is remained uncertain whether and how the cations would affect the NDMA formation kinetics in the river water.

Similar to what was observed in the lake water, the effects of cations in the river water could also be divided into the two general categories (i.e., “moderate to high” and “extremely high” hardness). To illustrate the two general sets of effects, as summarized in Figure 5-12, three
cation addition dosages (i.e., 0X, 0.5X, and 4X) were chosen for comparison about their impact on NDMA formation kinetics. The markers in the figure represent the measured NDMA molar conversions, while the curves are the estimated trends generated from the model (the same format was applied for Figure 5-13 and Figure 5-14).

![Graph](image)

**Figure 5-12.** NDMA molar conversions from ranitidine in Otonabee River water with 0X, 0.5X, and 4X cation additions (5 nM ranitidine, preformed monochloramine = 3.0 mg/L; error bars represent one standard deviation (n = 3))

The last measured point of the three conditions was all at 96 h. At 96h, with 0X and 4X cation additions, the NDMA formation was reaching the plateau, but with 0.5X addition, it seemed that a longer contact time is needed to achieve the ultimate NDMA formation. This observation somehow reflected that the reaction of ranitidine to form NDMA under 0.5X cation addition was the slowest among the three selected conditions. In fact, the Lag value of 0.5X condition was significantly different from that of the rest two conditions (p-values > 0.05; Tukey’s HSD test, 95 % confidence level). The higher Lag value of the 0.5X condition might be associated with its relatively longer initial lag period as shown in the figure since the rapid growing phase for all cases looked similar. It is interesting to see that the curves of no cation addition (0X) and 4X
addition looked alike; Tukey's HSD test was applied to compare the three model parameters from the two conditions (i.e., 0X and 4X), and the result suggested that the difference in the NDMA formation kinetics under these two conditions was insignificant (p-value > 0.05 for Lag, k, and θ; 95% confidence level). From Figure 5-12, it could be concluded that in Otonabee River water, initial addition of cations would slow down the NDMA formation from ranitidine, but further additions could result in a similar NDMA formation pattern as that of the original condition (without cation addition).

NDMA molar conversions from ranitidine in Otonabee River water with cation addition dosages from ambient levels (0X) to half their ambient levels (0.5X) are summarized in Figure 5-13.

![Figure 5-13](image)

**Figure 5-13.** NDMA molar conversions from ranitidine in Otonabee River water with 0X, 0.25X, and 0.5X cation additions (5 nM ranitidine, preformed monochloramine = 3.0 mg/L; error bars represent one standard deviation (n = 3))

As shown in the figure, the curves of 0X and 0.25X cation additions were almost overlapped, suggesting that the initial 0.25X cation addition (an increase in hardness from 111 to 157 mg/L as CaCO₃) to the river water did not result in a significant change in NDMA formation kinetics (p-value > 0.05 for θ, Lag, and k; Tukey's HSD test, 95% confidence level). Further addition of
0.25X (hardness level increased from 157 to 203 mg/L as CaCO₃) cations was found to result in a higher Lag value (p-value < 0.05; Tukey's HSD test, 95 % confidence level) with an observed longer initial lag period.

Since previous discussion suggested that after 0.5X cation addition, further addition of cations would tend to reduce the Lag value, the NDMA formation trends with addition dosages from 0.5X to 4X are summarized in Figure 5-14 for comparison. As shown in the figure, the length of the initial lag period reduced as the cation addition dosages increased. Roughly, the time required to achieve the ultimate NDMA formation was inversely related to the cation concentrations ranging from 0.5X to 4X. The rapid growing phase of 0.5X, X, and 2X were similar, which is in accordance with their similar k values. Therefore, changing the cations from 0.5X to 4X was found to affect the NDMA formation kinetics in terms of the Lag values.

![Figure 5-14. NDMA molar conversions from ranitidine in Otonabee River water with 0.5X, X, 2X, and 4X cation additions (5 nM ranitidine, preformed monochloramine = 3.0 mg/L; error bars represent one standard deviation (n = 3))](image-url)
5.3.4 Summary

To summarize, in Otonabee River water, the addition of cations did not affect the ultimate NDMA formation ($\theta$) and the pseudo-first reaction rate constant ($k$), but did affect the NDMA formation kinetics in terms of the model parameter $\text{Lag}$. The $\text{Lag}$ values first increased and gradually decreased as the cation concentrations increased. However, it has been noticed that the cation impact on NDMA formation in river water was minor; the most significant change was found associated with the addition of 0.5X cations.

5.4 Comparisons between Water Sources and to the Literature

Previous discussion in this thesis have suggested that cations affect the NDMA formation in both lake and river waters. It would be worthwhile to know whether there was any common trend or difference between the cation impacts on NDMA formation from ranitidine in the two water matrices. Therefore, the following discussion will compare the cation impact on each of the three model parameters in the two waters. To facilitate these comparisons and subsequent interpretations proposed in Section 5.5, the data were prepared and/or normalized in the following ways:

- They were examined relative to the total hardness in each test sample (Section 5.4.1)
- The hardness in each test sample was normalized to its DOC concentration (Section 5.4.2)
- The hardness in each test sample was normalized to the concentration of each NOM constituent (as determined by LC-OCD) (Section 5.4.3)

5.4.1 Hardness

Although the results from Chapter 4 have suggested that cations alone (in the absence of NOM) did not affect the NDMA formation from ranitidine, and since cations are positively charged and NOM usually contains negatively charged functional groups, cations have been found to bind with NOM to certain levels (Van Dijk, 1971), it is very likely that the observed cation impact on NDMA formation kinetics from ranitidine was a result of its interactions with NOM. It has been suggested that divalent cations, compared with monovalent cation, are more interactive with NOM by screening charges and reducing repulsion forces more effectively (Wang et al., 2013). Moreover, the interactions between the Ca$^{2+}$ and NOM were found to be the dominant factor
governing the NOM fouling of membranes (Lee et al., 2005). It is rational to believe that greater interactions between cations and NOM would result in a larger impact on NDMA formation from ranitidine, and as such, the presence of Ca\(^{2+}\) and Mg\(^{2+}\) might exert greater influence than Na\(^{+}\). Therefore, the total cation concentrations in the samples were calculated to their equivalent hardness levels (mg/L as CaCO\(_3\)) in the following discussion about their impact on NDMA formation from ranitidine. The estimated values of the model parameter \(\theta\) (ultimate NDMA formation) for both lake and river water at various hardness levels are summarized in Figure 5-15.

![Figure 5-15. Estimation of model parameter \(\theta\) for NDMA formation from ranitidine in lake and river water under various hardness levels (error bars represent one standard deviation \((n=3)\))](image)

For both waters, \(\theta\) remained relatively constant as the hardness increased; such similarity appeared to be enhanced as the hardness level increased. There was difference in \(\theta\) values of lake and river water when the hardness level was below 200 mg/L. However, the reason for this is remained unknown, and further preparation of additional samples at different cation concentrations within that range is beyond the scope of the current research. Nonetheless, NOM
has been suggested to have insignificant impact on the ultimate NDMA formation (Shen and Andrews, 2011b); if cations affect NDMA formation via their interactions with NOM, it is not likely that the cations would affect the ultimate NDMA formation.

The estimated values of the model parameter Lag for lake and river water under various hardness levels are summarized in Figure 5-16. It is very obvious that, under a similar hardness level, the Lag value of the river water was much higher than that of the lake water. This observation is in agreement with the findings from Shen (2013) which suggested that the Lag value was positively correlated with the TOC level. Also, in the present study, a similar correlation between the Lag value and the hardness level was found in both waters: as the hardness level increased, the Lag values for both waters first increased and subsequently decreased. The peak Lag values were observed at a hardness level of approximately 250 mg/L as CaCO₃ in both waters.

![Figure 5-16. Estimation of model parameter Lag for NDMA formation from ranitidine in lake and river water under various hardness levels (error bars represent one standard deviation (n=3))](image)

The estimated values of model parameter k (rate constant) for lake and river water under various hardness levels are summarized in Figure 5-17. It has been suggested that a higher TOC level
would tend to result in a lower k value of NDMA formation (Shen, 2013). In the present work, the pseudo-first reaction rate constant of NDMA formation in river water was much lower than that in lake water under a similar hardness level, confirming the result of Shen (2013). As the hardness level increased, the k values for NDMA formation in river water remained relatively constant, while the k values in lake water first decreased and later slightly increased. This observation might suggest that the k value was susceptible to the changes in hardness but only in Lake Ontario water (having a lower NOM concentration of 2.1 mg/L DOC relative to river water with 5.9 mg/L DOC).

![Figure 5-17](image)

**Figure 5-17. Estimation of model parameter k for NDMA formation from ranitidine in lake and river water under various hardness levels (error bars represent one standard deviation (n=3))**

### 5.4.2 Hardness/DOC Ratio

By normalizing the DOC and hardness levels into hardness/DOC ratios, the impact of cations on NDMA formation kinetics in lake and river waters could be more directly compared. The values of ultimate NDMA formation (θ) at various hardness/DOC ratios are summarized in Figure 5-18. The ultimate NDMA formation did not appear to be related to the hardness/DOC ratios. This is
as expected since cations were found not to affect the ultimate NDMA formation from ranitidine earlier in the present work.

Figure 5-18. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in lake and river water under various hardness/DOC ratios (error bars represent one standard deviation (n=3))

Figure 5-19 summarizes the Lag values of NDMA formation in both lake and river waters for the various hardness/DOC ratios. Both waters show an initial increase and a subsequent decrease in Lag. It was noticed that the Lag values of river water was very sensitive to the changes in hardness/DOC ratio within the ratio range tested (0 to 150). For example, the Lag value rise from 55.6 to 64.6 h as the hardness/DOC ratio increased from 19 to 35 and dropped from 64.6 to 49.7 h as the hardness/DOC ratio increased from 35 to approximately 144. While the Lag values of the lake water seemed only sensitive to the changes in Hardness/DOC ratio below the ratio of 114; as the ratio increased from 114 to 419, the Lag values of lake water gradually decreased from 16.6 to 10.8 h.
Figure 5-19. Estimation of model parameter Lag for NDMA formation from ranitidine in lake and river water under various hardness/DOC ratios (error bars represent one standard deviation (n=3)).

Figure 5-20 summarizes the k values for NDMA formation in both lake and river waters at various hardness/DOC ratios. The k values obtained in river water were found to be not sensitive to the changes in the Hardness/DOC ratio within the ratio range tested (19 to 144). In contrast, the k values for lake water samples appeared to be correlated to the Hardness/DOC ratio ranges: 0 to 100, 100 to 200, and 200 to above; within each ratio range, the k values were not statistically different. At a hardness/DOC ratio lower than 100, the observed k values were approximately one and a half times higher than at hardness/DOC ratios higher than 100. The lowest k value was observed in the hardness/DOC ratio range of 100 to 200, while approximately 15% higher k values were obtained as the hardness/DOC ratio increased to above 200.
5.4.3 Hardness/Humic Substances Ratio

Hardness concentrations were also normalized to the concentrations of selected NOM fractions for each sample as measured by LC-OCD in order to see if a particular fraction might be responsible for the observed cation effects. Although LC-OCD provides data for NOM fractions of LMW acids and neutrals, the current research focused on the fractions of humic substances, biopolymers, and building blocks as the most likely fractions to form complexes with cations in water. It turned out that all three NOM fractions of interest showed the same trends on the three model parameters (i.e., Lag, $\theta$, and $k$), so only those for the hardness/HS ratios are shown here, while the others (i.e., hardness/biopolymer and hardness/building blocks) are presented in Appendix 5.

Humic substances (HS) are major components of the natural organic matter (NOM) in water. As listed in Table 5-1, the HS fraction was 48.5 and 56.2 % of the DOC in lake and river water, respectively. Since HS molecules usually contain a lot of negatively charged functional groups
(i.e., carboxyl and phenolate groups), they have been found to interact with cations, especially divalent cations (i.e., Ca$^{2+}$ and Mg$^{2+}$) (Wang et al., 2013). A ratio of hardness/humic substances (hardness/HS) was used to quantify the relative magnitude of divalent cation (i.e., Ca$^{2+}$ and Mg$^{2+}$) and humic substances concentrations; the three model parameters (i.e., Lag, $\theta$, and k) under the various hardness/HS ratios tested were examined as follows.

The values of $\theta$ under various hardness/HS ratios in lake and river waters are summarized in Figure 5-21. Since the ultimate NDMA formation from ranitidine was found to be independent of the presence of cations in previous discussion, it was not surprising to see a similar non-effect here.

![Figure 5-21. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in lake and river water under various hardness/HS ratios (error bars represent one standard deviation (n=3))](image)

The Lag values at various hardness/HS ratios in lake and river waters are summarized in Figure 5-22. The Lag values for river water were monitored under hardness/HS ratio up to 260. In that ratio range, the Lag values for river water samples were so sensitive to the changes in hardness/HS ratio that it rapidly increased and then decreased. In a similar hardness/HS ratio
range, a quick increase of Lag values was observed in lake water samples. Further increase of hardness/HS ratio roughly from 250 to 910 in lake water resulted in a gradual decrease of the Lag values from 16.6 to 10.8 h.

![Figure 5-22. Estimation of model parameter Lag for NDMA formation from ranitidine in lake and river water under various hardness/HS ratios (error bars represent one standard deviation (n=3))](image)

The k values under various hardness/HS ratios in lake and river waters are summarized in Figure 5-23. Since the k values mostly describing the rapid increasing phase of NDMA formation, as shown in the figure, such phase of ranitidine to form NDMA was sensitive to the changes in hardness/HS ratio only in lake water. As the Hardness/HS ratio increased, the k values for river water samples were not statistically different, while that for lake water samples first decreased to a minimum at approximately 0.5X to X and subsequently increased with further increases in cations. Similar to the observation in k values under the various hardness/DOC ratios, the k values for lake water samples seemed to be correlated to the Hardness/HS ratio ranges: 0 to 200, 200 to 350, and 500 to above; within each ratio range, the k values were not statistically different.
Figure 5-23. Estimation of model parameter $k$ for NDMA formation from ranitidine in lake and river water under various hardness/HS ratios (error bars represent one standard deviation (n=3))

5.4.4 Summary

To summarize:

- The ultimate NDMA formation ($\theta$) from ranitidine was not susceptible to the changes in cation concentrations in both lake and river waters.
- The values for the rate constant parameter ($k$) were only susceptible to the changes in cation concentrations in lake water. As the cation concentrations increased, the $k$ values for lake water samples first decreased by approximately 65% and subsequently increased slightly (approximately 20 to 30%).
- Among the three model parameters, the Lag values were found most sensitive to the changes in cations in both waters, suggesting that cations primarily affect the initial formation of NDMA from ranitidine (lag period). Similar trends in the Lag parameter were observed in both waters: as the cation concentrations increased, the Lag values first increased and subsequently decreased.
• The cation impact was largely related to the presence of NOM, especially the fraction of HS.

• The Lag values for lake and river water samples were very sensitive to the changes in hardness/DOC or hardness/HS ratio within the low ratio range tested. For higher hardness/DOC or hardness/HS ratios, the Lag values were only monitored for lake water samples; they were found to gradually decrease as the two ratios were increased.

5.5 Possible Mechanisms for Cation Impacts

In general, the addition of cations was found to initially slow down and later accelerate the NDMA formation from ranitidine in the two selected natural waters. In both waters, the biggest fraction of NOM was the HS fraction (humic substances), large molecules with negatively charged functional groups, and therefore, they were suspected as the major NOM fraction which interacts with cations. In order to explain the cation impact, one must understand the interaction mechanism among cations, HS, and ranitidine.

5.5.1 Possible Interactions between HS and Ranitidine

The interaction mechanism between HS and ranitidine is largely unknown. Given that the pH in lake and river water samples were not higher than 8, the dimethylamine (DMA; pKₐ = 8.2) group of ranitidine would be largely protonated in both waters (66.6 % in lake water and 61.3 % in river water; calculated based on the conventional pKₐ-pH relationship: fraction of protonated ranitidine = \(1 \times \frac{1}{1+10^{(pK_a-pH)}}\)). The protonated DMA group (NH₃⁺) in the ranitidine molecule is likely to interact with the negatively charged groups (i.e., carboxyl and phenolic hydroxyl groups) in HS via electrostatic attraction. Several functional groups or atoms of the ranitidine molecule could potentially interact with HS via intermolecular attractions (e.g., van der Waals’ forces and hydrogen bonding). For example, the nitrogen and oxygen atoms in ranitidine could act as hydrogen bond acceptors, attracting hydrogen atoms from HS molecules. Similarly, the amino groups of ranitidine could form hydrogen bonds with the oxygen atoms in the HS molecules. In addition, a study concerning the interactions between ciprofloxacin and HS has suggested that HS would tend to rearrange its hydrophobic and hydrophilic regions and disrupt its intramolecular H-bonds to facilitate favorable intermolecular hydrogen bonding (Aristilde and Sposito, 2010). It is uncertain whether HS would undergo such conformational
changes when interacting with ranitidine, but it would be very likely that HS would have similar intermolecular interactions with ranitidine.

5.5.2 Possible Interactions between HS and Cations

It has been suggested that the interactions between divalent cations and HS include: (1) electrostatic attraction between cations and the negatively charged functional groups (i.e., carboxyl and phenolic hydroxyl groups) of HS; (2) cations as intra/intermolecular bridges connecting the carboxyl and phenolic hydroxyl groups of HS to form complexes (e.g., HS\(_1\)-cation\(^{2+}\)-HS\(_1\) and HS\(_1\)-cation\(^{2+}\)-HS\(_2\)) and leading to an increased aggregate size of HS (Wang et al., 2013). Either way, as the divalent cation concentrations are elevated, the negative charges of HS would tend to be neutralized, and the attractive intermolecular forces between two HS molecules would likely be increased (Brigante et al., 2007).

5.5.3 Possible Interactions between Cations and Ranitidine

The ranitidine molecule has been suggested to have a very strong coordinating ability, and could act as an effective ligand towards metal ions (e.g., Cu\(^{2+}\)) (Sovilj et al., 2003). Several functional groups or atoms in the ranitidine molecule have been found to form bonds with Cu\(^{2+}\), such as the sulfur, furan ring, 2-alkylamino-1-nitroethenic moiety, and the nitro group. The strongest attraction between ranitidine and Cu\(^{2+}\) was found to be through a chelating effect which is operative on the nitro group and the 2-alkylamino-1-nitroethenic moiety. Such interactions would result in a [Cu(ranitidine)\(_2\)]\(^{2+}\) complex as shown in Figure 5-24.

![Figure 5-24. Suggested structure of the [Cu(ranitidine)\(_2\)]\(^{2+}\) Complex (Sovilj et al., 2003)](image-url)
Moreover, as suggested by Ma and Dougherty (1997), cations (e.g., Na\(^+\)) and electron-rich π systems (e.g., furan ring) would tend to have cation-π interactions. Even though no studies to-date have investigated the interactions between ranitidine and the cations of interest in the present study, given that cations have been suggested to have ion-dipole interactions with the negative regions of neutral polar molecules (Myers, 2003), it is suspected that cations (i.e., Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^+\)) would interact with at least some of the atoms (e.g., sulfur, oxygen, and nitrogen) or functional groups (e.g., furan ring and nitro group) of ranitidine. It should be mentioned that such theories do not contradicted to the observation in Chapter 4 that cations alone do not affect the NDMA formation from ranitidine: those interactions between cations and ranitidine do not involve the DMA group of ranitidine, so the interactions between cations and ranitidine are unlikely to cause significant steric hindrance that could affect the reaction between its DMA group and chloramine to form NDMA.

5.5.4 Possible Interactions among HS, Ranitidine, and Cations

It was suggested in Section 5.5.3 that cations might have two types of interactions with ranitidine: intramolecular chelate effects and ion-dipole interactions. Given that cations (i.e., Ca\(^{2+}\) and Mg\(^{2+}\)) have been suggested to form complexes with HS (Section 5.5.2), it is suspected that HS-bound cations interacting with ranitidine could result in ternary ranitidine-cation-HS complexes via an intermolecular chelate formation or bridging effect. Even though there is no direct evidence to support such theory, a study by Aristilde and Sposito (2010) has revealed the existence of a ternary complex of ciprofloxacin-cation-HS. It would be expected that ranitidine-cation-HS complexes might be similar and lead to significant steric hindrance which could potentially slow down the reaction of NDMA formation from ranitidine. It should also be expected that as the aggregation level of HS becomes elevated by the addition of cations (as suggested in Section 5.5.2), the steric hindrance would become more significant. In another words, increasing the cation concentrations would slow down the NDMA formation from ranitidine.

As suggested in Section 5.5.1, ranitidine alone is likely to have two types (i.e., I and II) of interactions with HS:
I. Electrostatic attractions between the protonated DMA group (NH$_3^+$) of ranitidine and the negatively charged functional groups (i.e., carboxyl and phenolic hydroxyl groups) of HS:

\[ \text{NH}_3^+ \text{ of ranitidine} \xleftrightarrow{\text{electrostatic attraction}} \text{COO}^- \text{ and C}_6\text{H}_5\text{O}^- \text{ of HS} \]

II. Intermolecular attractions (hydrogen bonding) between ranitidine and HS

\[ \text{S, O, N atoms of ranitidine} \xleftrightarrow{\text{hydrogen bonding}} \text{H atom of HS} \]

\[ \text{H atom of ranitidine} \xleftrightarrow{\text{hydrogen bonding}} \text{O atom of HS} \]

Aristilde and Sposito (2010) suggested that the complexation of cations (e.g., Ca$^{2+}$, Mg$^{2+}$, and Fe(II)) appears to inhibit the binding of the positively charged amino group of ciprofloxacin with the negatively charged HS complexation sites. Similarly, it could be predicted that the interactions between cations and HS would impede the Type I interactions between ranitidine and HS. In addition, cations have been suggested to have ion-dipole interactions (stronger than hydrogen bonding) with hydrogen bond acceptors (i.e., S, O, and N atoms) from ranitidine and HS (Myers, 2003), which would potentially weaken Type II interactions between ranitidine and HS. Since the interactions between HS and ranitidine have been suggested to result in a slower formation of NDMA (Shen and Andrews, 2011b), if both Type I and II interactions are weakened then the presence of cations would tend to facilitate the NDMA formation from ranitidine.

5.5.5 Summary

As discussed above, cations could either inhibit or facilitate NDMA formation from ranitidine. Therefore, it is postulated that the initial addition of cations inhibited the initial NDMA formation from ranitidine by primarily enhancing the aggregation of HS and accordingly increasing the steric hindrance effect. As the aggregation level of HS reaches its maximum level, further addition of cations would start to accelerate the initial NDMA formation from ranitidine by changing the surface property of HS aggregates to weaken the electrostatic and intermolecular attractions (e.g., hydrogen bonding) between ranitidine and HS. All of these
interactions are summarized in Figure 5-25. Further research beyond the scope of the current thesis is needed to confirm these theories.

![Diagram of interactions between HS, ranitidine, and cations]

**Figure 5-25. Summary of possible interactions of HS, ranitidine, and cations**

### 5.6 Summary

In this chapter, NDMA formation from ranitidine in Lake Ontario water and Otonabee River water was monitored for 72 and 96 h, respectively, at six selected cation concentrations (i.e., 0X, 0.25X, 0.5X, X, 2X and 4X) representing an overall range of hardness levels of approximately 100 to 900 mg/L as CaCO₃. The kinetics of NDMA formation was characterized by applying a preexisting model, in which the formation kinetics was considered as a function of three model parameters (i.e., θ, Lag, and k).

The changes in cation concentrations did not affect the ultimate NDMA formation (θ) from ranitidine in both waters, but did affect the other NDMA formation kinetic parameters (i.e., Lag
and k). The parameter k was found susceptible to the changes in cations only in Lake Ontario water. In lake water, as the cation concentrations increased, the values of k first largely decreased and slightly increased. Unlike the parameter k, the parameter Lag was found susceptible to changes in cation concentrations in both waters. The degree of the cation impact on the parameter Lag was found related to the ratio of hardness/NOM (i.e., hardness/DOC, hardness/HS, hardness/biopolymers, and hardness/building blocks). The Lag values for lake and river water samples were very sensitive to the changes in hardness/NOM ratios within the low ratio range tested. At higher hardness/NOM ratios, the Lag values were monitored only for lake water samples and they were found to gradually decrease as the hardness/NOM ratio increased.

A possible mechanism of the cation impact on NDMA formation from ranitidine was postulated. In that mechanism, HS was suggested as the major NOM fraction which interacts with cations to affect the NDMA formation from ranitidine. The addition of cations was suggested to inhibit the initial NDMA formation from ranitidine by enhancing the steric hindrance; further addition of cations was suggested to accelerate the initial NDMA formation from ranitidine by changing the surface property of HS aggregates to weaken the electrostatic and intermolecular attractions between ranitidine and HS. These theories help lay the groundwork for future studies.
6 Conclusions and Recommendations

6.1 Summary

In this study, bench-scale experiments were conducted to investigate the impact of cation (i.e. Ca\(^{2+}\), Mg\(^{2+}\), and Na\(^{+}\)) concentrations on N-Nitrosodimethylamine (NDMA) formation from ranitidine in three water matrices (Milli-Q\(^{®}\) water, Lake Ontario water, and Otonabee River water) under practical chloramine disinfection conditions.

For experiments using Milli-Q\(^{®}\) water, NDMA formation was complete at 4 h. For experiments using lake and river water, NDMA formation kinetics was monitored for 72h and 96h, respectively. The kinetics of NDMA formation was characterized by applying a preexisting model, in which the formation kinetics was considered as a function of three model parameters: \(\theta\) (ultimate NDMA molar conversion); \(k\) (h\(^{-1}\)) (pseudo-first order reaction rate constant); Lag (h) (the time required to achieve 50 % of the ultimate NDMA molar conversion). Accordingly, the impact of cations on NDMA formation from ranitidine was discussed with respect to the changes in the magnitudes of the three model parameters.

6.2 Conclusions

The conclusions are summarized as the following:

- In Milli-Q\(^{®}\) water, the addition of excess cations did not change the yields of NDMA, suggesting that cations alone do not affect the NDMA formation from ranitidine.
- In lake and river water, elevating the cation concentrations also did not affect the ultimate NDMA formation (\(\theta\)), but it did affect the observed reaction rates for ranitidine to form NDMA:
  a. The reaction rates underwent an initial decrease and a subsequent increase as the cation concentration was increased.
  b. The changes in the observed rates were suggested to be primarily a result of the changes in the initial lag period of NDMA formation under various cation concentrations.
- A possible mechanism of the cation impact on NDMA formation from ranitidine was postulated:
a. The addition of cations initially enhances the aggregation of humic substances (HS) in the two natural waters tested, resulting in higher steric hindrance on ranitidine, thus inhibiting the formation of NDMA from “free” ranitidine.

b. Further increases in cation concentration appear to modify the surface property of HS, resulting in less electrostatic or intermolecular attraction between ranitidine and HS, which thus facilitates the increase in observed rates of NDMA formation.

6.3 Recommendations for Future Work

- In the present study, the impact of cations on NDMA formation from ranitidine has been postulated to be associated with the possible interactions among cations, NOM and ranitidine. In order to better understand the mechanism of such interactions, synthetic water samples could be used to further investigate the cation impact on NDMA formation from ranitidine in the presence of NOM.

- Cations have been found to affect the NDMA formation from ranitidine in the two selected natural waters. However, due to the relatively rare occurrence of ranitidine in source waters, it is unlikely that ranitidine would contribute the majority formation of NDMA upon chloramination in distribution systems. It would be worthwhile to study the cation impact on NDMA formation from other known precursors.
References


AWWA, 2006. Formation and decay of disinfection by-products in the distribution system.


California Department of Public Health, 2011. A brief history of NDMA findings in drinking water.


Office of Environmental Health Hazard Assessment (OEHHA), 2006. Public Health Goal for N-nitrosodimethylamine and cadmium in drinking water.


Sadmani, A.H.M.A., 2013. Personal communication, Department of Civil Engineering, University of Toronto.


Appendix 1  24 h Chloramine Demand Determination

24 h chloramine demand was determined in the two selected natural waters: Lake Ontario water and Otonabee River water. Three initial monochloramine dosages (2.5, 5, and 10 mg/L as total Cl\textsubscript{2}) were applied to each water type, and samples were prepared in duplicate. The total chlorine concentration in each sample was measured after 24 h (24 h total chlorine residual). The raw data were shown in Table A1-1 and Table A1-2, respectively, for Lake Ontario water and Otonabee River water. A linear regression was applied between the actual monochloramine dosage (as total Cl\textsubscript{2}) and the 24 h total chlorine residual for each water as shown in Figure A1- and Figure A1-. The chloramine dosage (2.5 mg/L + 24 h demand) for NDMA formation test was determined by calculating the respective chloramine dosage for the 24 h total chlorine residual of 2.5 mg/L using the linear regression line equations. Thus, the chloramine dosage for NDMA formation test in Lake Ontario water and Otonabee River water has been determined as 2.8 and 3.0 mg/L, respectively.

Table A1-1. Raw data of the 24 h chloramine demand test in Lake Ontario water

<table>
<thead>
<tr>
<th>Target monochloramine dosage as total Cl\textsubscript{2} (mg/L)</th>
<th>Actual monochloramine dosage as total Cl\textsubscript{2} (mg/L)</th>
<th>24 h total Cl\textsubscript{2} residual (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.5</td>
<td>2.45</td>
<td>2.28</td>
</tr>
<tr>
<td>5</td>
<td>4.91</td>
<td>4.53</td>
</tr>
<tr>
<td>10</td>
<td>9.81</td>
<td>8.85</td>
</tr>
</tbody>
</table>

Table A1-2. Raw data of the 24 h chloramine demand test in Otonabee River water

<table>
<thead>
<tr>
<th>Target monochloramine dosage as total Cl\textsubscript{2} (mg/L)</th>
<th>Actual monochloramine dosage as total Cl\textsubscript{2} (mg/L)</th>
<th>24 h total Cl\textsubscript{2} residual (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.5</td>
<td>2.5</td>
<td>1.99</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>4.48</td>
</tr>
<tr>
<td>10</td>
<td>10</td>
<td>9.825</td>
</tr>
</tbody>
</table>
Figure A1-1. 24 h total Cl₂ residual under various monochloramine dosages in lake water

\[ y = 0.8918x + 0.1125 \]
\[ R^2 = 0.9999 \]

Figure A1-2. 24 h total Cl₂ residual under various monochloramine dosages in river water

\[ y = 1.0483x - 0.685 \]
\[ R^2 = 0.9997 \]
Appendix 2  Calibration Curves for NDMA

For each of the three waters (i.e., Milli-Q® water, Lake Ontario water, and Otonabee River water), a 6-level calibration curve was prepared to calculate the NDMA concentration in the chloraminated samples as given in Figure A2-1, Figure A2-2, and Figure A2-3.

![Calibration Curve for NDMA Concentration in Milli-Q® Water Samples](image)

**Figure A2-1.** Calibration curve for NDMA concentration in Milli-Q® water samples
Figure A2-2. Calibration curve for NDMA concentration in lake water samples

\[ y = 0.0131x + 0.0114 \]
\[ R^2 = 0.9844 \]

Figure A2-3. Calibration curve for NDMA concentration in river water samples

\[ y = 0.0159x \]
\[ R^2 = 0.9797 \]
\[ y = 0.0231x - 1.1376 \]
\[ R^2 = 0.9809 \]
Appendix 3  Raw Data for NDMA Formation in The Three Water Matrices

NDMA formation in the three water matrices is summarized in the following tables.

### Table A3-1. NDMA formation in Milli-Q® water samples

<table>
<thead>
<tr>
<th></th>
<th>0X</th>
<th>X</th>
<th>2X</th>
<th>5X</th>
</tr>
</thead>
<tbody>
<tr>
<td>NDMA concentration in blank control samples (ng/L)</td>
<td>25.2 (4.2)*</td>
<td>24.0 (4.0)</td>
<td>26.7 (2.3)</td>
<td>22.5 (2.8)</td>
</tr>
<tr>
<td>NDMA concentration in triplicate test samples (ng/L)</td>
<td>88.2 (11.4)</td>
<td>82.5 (19.5)</td>
<td>105 (13.3)</td>
<td>82.5 (17.7)</td>
</tr>
<tr>
<td>NDMA molar conversion in triplicate test samples</td>
<td>23.8 % (3.1 %)</td>
<td>22.3 % (5.3 %)</td>
<td>28.3 % (3.6 %)</td>
<td>22.3 % (4.8 %)</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca²⁺, 20 mg/L Mg²⁺, and 10 mg/L Na⁺ of added cations
* Numbers in these brackets represent the standard deviation of each parameter

### Table A3-2. NDMA formation in Lake Ontario water samples

<table>
<thead>
<tr>
<th>Contact time (h)</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>5.3 (0.6)*</td>
<td>N/A</td>
<td>N/A</td>
<td>3.7 (0.5)</td>
<td>N/A</td>
<td>3.8 (0.5)</td>
</tr>
<tr>
<td>4</td>
<td>42.4 (2.4)</td>
<td>2.6 (2.3)</td>
<td>0 (2.2)</td>
<td>16.8 (3.7)</td>
<td>9.8 (7.6)</td>
<td>26.6 (6.1)</td>
</tr>
<tr>
<td>8</td>
<td>150 (4.4)</td>
<td>76.8 (6.7)</td>
<td>45.8 (5.2)</td>
<td>96.2 (6.7)</td>
<td>45.4 (9.6)</td>
<td>127 (9.8)</td>
</tr>
<tr>
<td>12</td>
<td>255 (16.3)</td>
<td>274 (44.2)</td>
<td>276 (19.9)</td>
<td>140 (7.8)</td>
<td>243 (35.7)</td>
<td>175 (14.8)</td>
</tr>
<tr>
<td>16</td>
<td>287 (34.2)</td>
<td>273 (10.7)</td>
<td>176 (11.8)</td>
<td>168 (19.6)</td>
<td>189 (18.9)</td>
<td>212 (8.2)</td>
</tr>
<tr>
<td>24</td>
<td>294 (8.0)</td>
<td>319 (10.7)</td>
<td>201 (41.9)</td>
<td>198 (6.6)</td>
<td>228 (38.0)</td>
<td>256 (21.8)</td>
</tr>
<tr>
<td>48</td>
<td>313 (11.2)</td>
<td>305 (24.6)</td>
<td>310 (14.1)</td>
<td>266 (2.0)</td>
<td>296 (37.1)</td>
<td>286 (10.3)</td>
</tr>
<tr>
<td>72</td>
<td>316 (12.9)</td>
<td>261 (3.2)</td>
<td>292 (9.2)</td>
<td>313 (5.5)</td>
<td>282 (3.5)</td>
<td>318 (10.3)</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca²⁺, 20 mg/L Mg²⁺, and 10 mg/L Na⁺ of added cations
* Numbers in these brackets represent the standard deviation of each parameter
<table>
<thead>
<tr>
<th>Contact time (h)</th>
<th>0X</th>
<th>0.25X</th>
<th>0.5X</th>
<th>X</th>
<th>2X</th>
<th>4X</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td>10.2 (3.7)*</td>
<td>4.8 (1.4)</td>
<td>4.0 (3.2)</td>
<td>9.4 (2.2)</td>
<td>13.4 (0.4)</td>
<td>12.0 (1.5)</td>
</tr>
<tr>
<td>16</td>
<td>15.6 (1.1)</td>
<td>13.9 (4.3)</td>
<td>12.0 (0.5)</td>
<td>16.6 (6.3)</td>
<td>19.0 (12.9)</td>
<td>18.6 (4.3)</td>
</tr>
<tr>
<td>24</td>
<td>42.6 (10.6)</td>
<td>38.5 (8.8)</td>
<td>23.3 (7.3)</td>
<td>35.2 (2.1)</td>
<td>42.2 (1.8)</td>
<td>49.2 (1.7)</td>
</tr>
<tr>
<td>48</td>
<td>77.4 (8.4)</td>
<td>73.7 (4.5)</td>
<td>73.9 (7.8)</td>
<td>76.8 (5.7)</td>
<td>88.2 (6.9)</td>
<td>127 (2.7)</td>
</tr>
<tr>
<td>72</td>
<td>216 (0.6)</td>
<td>209 (13.7)</td>
<td>178 (10.9)</td>
<td>188 (11.9)</td>
<td>199 (8.0)</td>
<td>242 (38.9)</td>
</tr>
<tr>
<td>96</td>
<td>242 (10.4)</td>
<td>240 (8.9)</td>
<td>255 (32.2)</td>
<td>249 (38.9)</td>
<td>246 (28.8)</td>
<td>269 (18.9)</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations

* Numbers in these brackets represent the standard deviation of each parameter
Appendix 4  Statistical Analysis Results

All of the statistical analyses were performed using the GraphPad Prism® ANOVA tool.

For experiments with Milli-Q® Water, One-way ANOVA tests were performed for the blank control and triplicate (with ranitidine) samples, respectively. The tests (one factor: cation concentration; four-level: cation addition dosage of 0X, X, 2X, and 5X) were to determine if the cation impact was significant on the NDMA formation, and the test results are summarized in Table A4-1.

Table A4-1. Summary of the one-way ANOVA results (95 % confidence level) for Milli-Q® water experiments

<table>
<thead>
<tr>
<th>Test target</th>
<th>P value</th>
<th>Are means significant different? (P &lt; 0.05)</th>
<th>Number of groups</th>
<th>F</th>
<th>R²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blank control samples</td>
<td>0.5203</td>
<td>No</td>
<td>4</td>
<td>0.82</td>
<td>0.2343</td>
</tr>
<tr>
<td>Triplicate test samples (with ranitidine)</td>
<td>0.3454</td>
<td>No</td>
<td>4</td>
<td>1.28</td>
<td>0.3243</td>
</tr>
</tbody>
</table>

For experiments with Lake Ontario water and Otonabee River water, a one-way ANOVA test was performed for each NDMA formation kinetics model parameter (i.e., Lag, θ, and k) followed by a Tukey’s HSD (honestly significant difference) test. The one-way ANOVA (one factor: cation concentration; six-level: cation addition dosage of 0X, 0.25X, 0.5X, X, 2X, and 4X) test was to determine if the cation impact was significant, while the Tukey’s HSD test was to determine how significant the difference was between each pair of cation dosages applied.
Table A4-2. Summary of the one-way ANOVA and Tukey’s HSD results (95 % confidence level) for Lake Ontario water experiments

<table>
<thead>
<tr>
<th>Test</th>
<th>Pair comparison</th>
<th>Significant? (P&lt;0.05?)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0X vs 0.25X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0X vs 0.5X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0X vs X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0X vs 2X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.25X vs 0.5X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.25X vs X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.25X vs 2X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Tukey's HSD Test</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25X vs 4X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0.5X vs X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0.5X vs 2X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.5X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>X vs 2X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>2X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^+$ of added cations
Table A4-3. Summary of the one-way ANOVA and Tukey’s HSD results (95% confidence level) for Otonabee River water experiments

<table>
<thead>
<tr>
<th>Test</th>
<th>0</th>
<th>Lag (h)</th>
<th>k</th>
</tr>
</thead>
<tbody>
<tr>
<td>P value</td>
<td>0.0821</td>
<td>0.0003</td>
<td>0.0508</td>
</tr>
<tr>
<td>Are means significantly different? (P&lt;0.05)</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>One-Way ANOVA</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of groups</td>
<td>6</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>F</td>
<td>2.59</td>
<td>11.71</td>
<td>3.089</td>
</tr>
<tr>
<td>$R^2$</td>
<td>0.519</td>
<td>0.83</td>
<td>0.5628</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Test</th>
<th>Pair comparison</th>
<th>Significant? (P&lt;0.05?)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0X vs 0.25X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0X vs 0.5X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0X vs X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0X vs 2X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0X vs 4X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0.25X vs 0.5X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.25X vs X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>0.5X vs 2X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>0.5X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>X vs 2X</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>2X vs 4X</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>

X = 40 mg/L Ca$^{2+}$, 20 mg/L Mg$^{2+}$, and 10 mg/L Na$^{+}$ of added cations
Appendix 5  Kinetic Model Parameters at Various Hardness/NOM Fraction Ratios

Figure A5-1. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in lake and river water under various hardness/biopolymers ratios (error bars represent one standard deviation (n=3))
Figure A5-2. Estimation of model parameter Lag for NDMA formation from ranitidine in lake and river water under various hardness/biopolymers ratios (error bars represent one standard deviation (n=3)).
Figure A5-3. Estimation of model parameter $k$ for NDMA formation from ranitidine in lake and river water under various hardness/biopolymers ratios (error bars represent one standard deviation ($n=3$)).
Figure A5-4. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in lake and river water under various hardness/building blocks ratios (error bars represent one standard deviation ($n=3$))
Figure A5-5. Estimation of model parameter Lag for NDMA formation from ranitidine in lake and river water under various hardness/building blocks ratios (error bars represent one standard deviation (n=3))
Figure A5-6. Estimation of model parameter $\theta$ for NDMA formation from ranitidine in lake and river water under various hardness/building blocks ratios (error bars represent one standard deviation ($n=3$)).