ACTIVATED CARBON CATALYZED NITROSAMINE FORMATION VIA AMINE NITROSATION

A Thesis
Presented to
The Academic Faculty

by

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Master of Science in the
School of Civil and Environmental Engineering

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ACTIVATED CARBON CATALYZED NITROSAMINE
FORMATION VIA AMINE NITROSATION

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<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
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<tr>
<td>AC</td>
<td>Activated carbon</td>
</tr>
<tr>
<td>AqC</td>
<td>Siemens AquaCarb 1230C Granular Activated Carbon</td>
</tr>
<tr>
<td>CCL3</td>
<td>Contaminant Candidate List 3</td>
</tr>
<tr>
<td>DBA</td>
<td>Dibutylamine</td>
</tr>
<tr>
<td>DBPs</td>
<td>Disinfection byproducts</td>
</tr>
<tr>
<td>DCM</td>
<td>Dichloromethane</td>
</tr>
<tr>
<td>DEA</td>
<td>Diethylamine</td>
</tr>
<tr>
<td>DMA</td>
<td>Dimethylamine</td>
</tr>
<tr>
<td>DMAFuOH</td>
<td>5-dimethylaminofurfurylalcohol</td>
</tr>
<tr>
<td>DMBzA</td>
<td>Dimethylbenzylamine</td>
</tr>
<tr>
<td>EPA</td>
<td>United States Environmental Protection Agency</td>
</tr>
<tr>
<td>GAC</td>
<td>Granular activated carbon</td>
</tr>
<tr>
<td>LLE</td>
<td>Liquid-liquid extraction</td>
</tr>
<tr>
<td>MOR</td>
<td>Morpholine</td>
</tr>
<tr>
<td>MW</td>
<td>Molecular weight</td>
</tr>
<tr>
<td>NanoAg</td>
<td>Nanosilver particles</td>
</tr>
<tr>
<td>NDBA</td>
<td>N-Nitrosodi-n-butylamine</td>
</tr>
<tr>
<td>NDEA</td>
<td>N-Nitrosodiethylamine</td>
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<td>NDMA</td>
<td>N-Nitrosodimethylamine</td>
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<td>NDMA-d6</td>
<td>N-Nitrosodimethylamine-d6</td>
</tr>
<tr>
<td>NDPA</td>
<td>N-Nitrosodi-n-propylamine</td>
</tr>
<tr>
<td>NDPhA</td>
<td>N-Nitrosodiphenylamine</td>
</tr>
<tr>
<td>NMA</td>
<td>N-methylaniline</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Description</td>
</tr>
<tr>
<td>--------------</td>
<td>----------------------------------</td>
</tr>
<tr>
<td>NMEA</td>
<td>N-Nitrosomethylethylamine</td>
</tr>
<tr>
<td>NMOR</td>
<td>N-Nitrosomorpholine</td>
</tr>
<tr>
<td>NNMA</td>
<td>N-Nitroso-n-methylaniline</td>
</tr>
<tr>
<td>NO$_2^-$</td>
<td>Nitrite</td>
</tr>
<tr>
<td>NO$_3^-$</td>
<td>Nitrate</td>
</tr>
<tr>
<td>NPIP</td>
<td>N-Nitrosopiperidine</td>
</tr>
<tr>
<td>NPYR</td>
<td>N-Nitrosopyrrolidine</td>
</tr>
<tr>
<td>PAC</td>
<td>Powdered activated carbon</td>
</tr>
<tr>
<td>pH$_{PZC}$</td>
<td>Point of zero charge</td>
</tr>
<tr>
<td>PYR</td>
<td>Pyrrolidine</td>
</tr>
<tr>
<td>RAN</td>
<td>Ranitidine</td>
</tr>
<tr>
<td>SPE</td>
<td>Solid phase extraction</td>
</tr>
<tr>
<td>TBA</td>
<td>Tert-butyl alcohol</td>
</tr>
<tr>
<td>TMA</td>
<td>Trimethylamine</td>
</tr>
<tr>
<td>UCMR2</td>
<td>Unregulated Contaminant Monitoring Rule 2</td>
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SUMMARY

Nitrosamines have garnered increasing attention from researchers and policy makers in recent years due to potential human health implications associated with their unintentional formation in water and wastewater treatment facilities. This work addresses a crucial nitrosamine formation pathway concerning the catalysis of amine nitrosation by activated carbon materials whose use is widespread in municipal and industrial systems. Experimental results show that this catalysis is highly pH dependent, with maximum formation achieved near the pKa value for each of the secondary amines tested. This result suggests that the overall formation potential is governed by individual amine properties and their interactions with carbon surfaces, rather than solely nitrite speciation as previously reported. Formation of the most commonly studied nitrosamine, N-nitrosodimethylamine, was shown to be highly dependent on initial dimethylamine (DMA) solution concentration, with yields of approximately 0.11% of the spiked secondary amine at pH 7.5. Morpholine and dibutylamine, larger and bulkier secondary amines, formed their corresponding nitrosamines at higher yields than DMA (0.21% and 1.69%, respectively). Additionally, select tertiary amines were shown to be capable of undergoing nitrosation on the same order of magnitude as the secondary amines under neutral conditions in the presence of activated carbons. The magnitude of these results indicates that greater attention should be paid to this previously overlooked mechanism for nitrosamine formation.
CHAPTER 1
INTRODUCTION

*N*-nitrosamines are among a class of emerging contaminants known as nitrogenous disinfection byproducts. Although they are not currently subject to any federal regulations, nitrosamines may be unintentionally formed in water and wastewater treatment facilities and lead to a significant public health risk due to their potentially carcinogenic properties (1, 2). They are currently being considered for regulation under the United States Environmental Protection Agency (EPA) Contaminant Candidate List 3 (CCL3) (3) and Unregulated Contaminant Monitoring Rule 2 (UCMR2) (4). In recent years, attempts to minimize the presence of regulated disinfection byproducts (DBPs) such as trihalomethanes and haloacetic acids have resulted in a surge in the use of chloramines as an alternative to traditional chlorination (5). Since chloramines have been shown to dramatically enhance nitrosamine formation (6), this trend will likely exacerbate the current issues surrounding nitrosamine detection. Other water disinfection practices including ozonation, free chlorination, and combined UV/chlorine have also been shown to produce nitrosamines (7, 8, 9), although typically at a lower yield than chloramination.

*N*-Nitrosodimethylamine (NDMA) is the most commonly studied nitrosamine, with levels above 2 ng/L in 10% of water samples collected from public drinking water treatment systems nationwide and reported concentrations reaching 630 ng/L in North America based on the survey results of UCMR2 (10). The molecular structures, corresponding cancer risk concentrations, and reported detected drinking water concentrations in China for nitrosamines listed in the CCL3 and UCMR2 are summarized in Table 1.
### Table 1. Nitrosamine Species Information.

<table>
<thead>
<tr>
<th>Nitrosamine Species</th>
<th>Symbol</th>
<th>MW (g/mol)</th>
<th>Structure</th>
<th>Reported Concentration (ng/L) (11)</th>
<th>10⁻⁶ Cancer Risk Concentration (ng/L) (1)</th>
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<tr>
<td>N-Nitrosodimethylamine</td>
<td>NDMA</td>
<td>74.1</td>
<td><img src="image1.png" alt="Structure" /></td>
<td>3.0 – 36</td>
<td>0.7</td>
</tr>
<tr>
<td>N-Nitroso-n-methylethylamine</td>
<td>NMEA</td>
<td>88.1</td>
<td><img src="image2.png" alt="Structure" /></td>
<td>2.9 – 4.5</td>
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<tr>
<td>N-Nitrosopyrrolidine</td>
<td>NPYR</td>
<td>100.1</td>
<td><img src="image3.png" alt="Structure" /></td>
<td>2.4 – 13</td>
<td>20</td>
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<td>N-Nitrosodiethylamine</td>
<td>NDEA</td>
<td>102.1</td>
<td><img src="image4.png" alt="Structure" /></td>
<td>6.4 – 19.5</td>
<td>0.2</td>
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<tr>
<td>N-Nitrosopiperidine</td>
<td>NPIP</td>
<td>114.2</td>
<td><img src="image5.png" alt="Structure" /></td>
<td>4.1 – 23</td>
<td>3.5</td>
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<td>N-Nitrosomorpholine</td>
<td>NMOR</td>
<td>116.1</td>
<td><img src="image6.png" alt="Structure" /></td>
<td>2.5 – 12</td>
<td>5</td>
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<td>N-Nitrosodi-n-propylamine</td>
<td>NDPA</td>
<td>130.1</td>
<td><img src="image7.png" alt="Structure" /></td>
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<td>N-Nitroso-n-methylaniline</td>
<td>NNMA</td>
<td>136.2</td>
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<td>N-Nitrosodi-n-butylamine</td>
<td>NDBA</td>
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<td>N-Nitrosodiphenylamine</td>
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<td><img src="image10.png" alt="Structure" /></td>
<td>ND – 6.0</td>
<td>7,000</td>
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The conversion of organic molecules into those containing the nitroso functional group (R-N=O) is known as nitrosation. This process can occur with amines as well as amides (12) and may target sulfur, as in S-nitrosation (13), or nitrogen, as in N-nitrosation (14). Both secondary and tertiary amines have been shown to undergo N-nitrosation, while primary amines are not expected to yield appreciable amounts of nitrosamines since they are relatively unstable in water (15, 16, 17). Some common
nitrosating agents include NO$_3^-$, NO$_2^-$, NO, N$_2$O$_4$, and N$_2$O$_3$ (15, 18). The widespread use of nitrate and nitrite in the preservation of cured meats over the last several decades has caused a surge in research due to the likelihood of nitrosation to occur inside the stomach and generate suspected cancer-causing compounds.

While a great deal of attention has been paid to the formation of nitrosamines via nitrosation of secondary amines within the human gastrointestinal tract, the relevance of this phenomenon to water and wastewater treatment has been largely overlooked since it was thought that nitrosation could only occur in acidic environments (19, 20). This hypothesis was based on the presumption that nitrite must first be converted to nitrous acid (pKa = 3.4) before attacking the lone pair of electrons present on an amine’s nitrogen atom (18).

However, a number of studies have shown that certain materials have the ability to augment the transformation of amine precursors into nitrosamines under relatively neutral pH conditions relevant in water treatment and distribution systems (11). Some studies have provided evidence that nitrosation can be facilitated in neutral pH conditions when in the presence of chemical catalysts such as fulvic acid, formaldehyde (21, 22), and activated carbon (AC) (23, 24, 25). In particular, the capacity of AC to catalyze the formation of nitrosamines is concerning due to the widespread use of such materials as adsorbents in polishing steps for water and wastewater treatment (25). Water treatment applications have been estimated to account for approximately 44% of the more than 500 million pounds of AC produced annually (26), while more recent market research places total AC demand around 1.3 billion pounds per year by 2017 (27). Furthermore, this catalysis represents a potential source of analytical error since the primary method for measuring nitrosamines in aqueous samples, EPA Method 521, utilizes AC as a solid phase extraction (SPE) adsorbent (28).

The primary goal of this study was to highlight previously unknown aspects of AC catalyzed nitrosamine formation. While previous studies (24, 25) have illuminated
details of AC’s effects to catalyze oxidation of amines adsorbed on carbon surfaces with atmospheric nitrogen and oxygen to generate nitrosamines, little research focus has thus far been placed on catalyzed nitrosation of amines with nitrite by carbon materials. Results from limited previous research (23) suggest a strong pH dependence of AC-catalyzed nitrosation with higher nitrosamine formation under more acidic conditions, but only a very limited range of conditions were investigated. Furthermore, this study aimed to shed new light on the relative importance of various precursor sources, environmental conditions, and potential carbon pretreatment measures that may influence nitrosamine generation via the AC-catalyzed nitrosation, thereby providing potential strategies to lessen undesirable nitrosamine formation.
CHAPTER 2
MATERIALS AND METHODS

2.1. Chemicals

Standard grade NDMA, NPYR, NDEA, NMOR, NDBA, NDMA, methylene chloride, trimethylamine (TMA) hydrochloride, dimethylamine hydrochloride, dibutylamine (DBA), pyrrolidine (PYR), diethylamine (DEA), morpholine (MOR), dimethylbenzylamine (DMBzA), ranitidine (RAN), 5-dimethylaminomethylfurfuryl alcohol (DMAFuOH), monobasic and dibasic sodium phosphate, sodium nitrite, sodium nitrate, hydrochloric acid, sulfuric acid, benzenesulfonyl chloride, acetone, nitric acid, sodium borate were purchased at >98% purity from Fisher Scientific or Sigma-Aldrich. Dimethylamine-d6 and NDMA-d6 were obtained from Cambridge Isotope Laboratories. Granular activated carbon materials were obtained from Siemens (AqC 1230C) and powdered carbons were provided by Norit (PAC 200). Carbon cartridges were purchased from United Chemical Technologies (UCT Enviro Clean Extraction Columns). Reagents for nitrite and nitrate measurements were purchased from Hach (NitriVer3 and NitraVer5). No further purification was done for any reagents. Deionized (DI) water was produced by a Millipore Milli-Q® water purification system.

2.2. Experimental Setup

2.2.1. Granular Activated Carbon (GAC) Experiments

Unless otherwise noted, 100 mg carbon was added to 250 mL of solution containing 50 µM amine, 50 µM nitrite, 10 mM phosphate buffer at pH 7.5, and 31.25 nM NDMA-d6 as an internal standard. Spiking concentrations were chosen based on
values that might be found in typical wastewater treatment facilities. The EPA has set maximum contaminant levels of 10 mg/L NO$_3^-$-N and 1 mg/L NO$_2^-$-N for nitrate and nitrite (161 µM and 22 µM), while DMA concentrations range from 20-1,000 µg/L (0.44-22 µM) (2, 29, 30, 31). For experiments utilizing real wastewater samples, aqueous samples were collected from the secondary effluent of a local municipal wastewater treatment plant and filtered through 1.0 µm glass fiber filters without adjusting pH prior to solution preparation. The resulting solutions, prepared in triplicate, were shaken for 2 hours, filtered through glass fiber filters and the filtrate was set aside for further amine and nitrite analysis. Carbon particles were collected from the filters, placed in microcentrifuge tubes, mixed with 1.5 mL dichloromethane (DCM), and centrifuged at 5,000 RPM for 5 minutes. The solvent layers were then transferred to GC vials and analyzed for nitrosamines. Samples prepared without AC were subjected to liquid-liquid extraction (LLE) using 10 mL DCM instead of the typical filtration and carbon collection method.

2.2.2. Powdered Activated Carbon (PAC) Experiments

Since powdered activated carbon (PAC) is difficult to recover upon filtration due to its adhesive properties on moist surfaces, a separate protocol had to be developed for analysis of fine carbon materials. Samples were prepared in 50 mL centrifuge tubes using the previously detailed precursor concentrations and carbon loadings. The mixtures were allowed to react for 2 hours and then spiked with 2 mL DCM to elute the analytes into the liquid phase before filtration through 1 µm glass fiber filters. The filtrate was then injected with an additional 5 mL DCM to increase solvent volume for ease of extraction before transferring the solvent layer to GC vials for measurement. Results were corrected for NDMA-d6 extraction efficiency to ensure consistency with other methods.
2.2.3. Reactions with AC Cartridges

Studies employing the AC cartridge method required cartridge conditioning prior to sample filtration, as detailed in the EPA Method 521 (28). For this process, 6 mL DCM was passed through the cartridges followed by 6 mL methanol, and finally 15 mL DI water. Cartridges were not allowed to dry completely after the methanol and water were filtered through. Aqueous samples were passed through the conditioned cartridges at a rate of 5-10 mL/min using a Thermo Scientific 24 port manifold system. The filtrate from this process was collected and stored for later secondary amine and nitrite adsorption analysis. Analytes were then eluted from the carbon surfaces by soaking in 3 mL DCM for 1-2 minutes before the solvent was collected in amber vials. This process was repeated to generate a final DCM volume of 10 mL prior to analysis with GC-MS. The AC particles inside the AC cartridges were also tested by the GAC particle method for a direct comparison with the cartridge method. In this case, the cartridges were subjected to identical pre-conditioning processes before the AC was removed from the cartridges and added to the amber bottle batch reactors. In addition to DI water matrices, real water samples were used in these studies. In this case, the real water samples were measured for nitrite (NO$_2^-$) and dimethylamine (DMA) concentrations initially and spiked to a total concentration of 50 µM for each constituent when appropriate.

2.3. Nitrosamine and Precursor Analyses

Nitrosamine analysis was conducted similarly to the protocol described previously (25). All experimental results were corrected for surrogate (NDMA-d6) extraction efficiencies of approximately 20%. The relevant GC-MS conditions and monitoring ions are shown in Appendix A.

Secondary amines were analyzed by extracting 8.8 mL samples from the aforementioned filtrate. The samples were spiked with 1.96 µM DMA-d6 as an internal
standard, 1 mM benzenesulfonyl chloride as a derivatizing agent, and adjusted to pH 9.0 with 10 mM borate buffer for a final volume of 10 mL. Samples were then shaken for 2 hours at room temperature before spiking with 2 mL DCM for LLE. After DCM addition, the mixtures were shaken for 5 minutes before the solvent layers were transferred to GC vials and processed by GC-MS. Results were normalized by derivatization efficiency, which averaged around 80%.

Nitrite was measured by collecting 10 mL volumes from the filtrate and mixing with Hach reagent powders according to their established protocol. Samples were shaken for 20 minutes and absorbance was then measured on a visible light spectrophotometer at a wavelength of 507 nm. Both nitrite and amine adsorption were calculated by subtracting the final measured concentrations from the initial spiked values.

2.4. AC Modification

Protocols used to modify the surfaces of AC particles were adopted from recent literature (25). Siemens AqC carbon was used in all modified AC samples. For acid treatment, 2 g of particles were submerged in 50 mL of concentrated HNO₃ or H₂SO₄ and shaken for 30 minutes prior to rinsing with DI water for 2 minutes. During rinsing, particles were shaken with clean DI water, drained, and then shaken with another fresh batch of water to minimize residual chemicals in the AC pores before air drying. Similar procedures were followed for treatment with tert-butyl alcohol. For heat treatment, particles were weighed to 2 grams, heated at 600 °C for 6 hours, and cooled to room temperature before use in the experiments. Nano-silver doped Siemens AqC was prepared by Dr. Martina Karlikova at the Palacky University, Olomouc, Czech Republic, by adsorption of silver nanoparticles (average size ~ 30 nm) onto the AqC particles.
CHAPTER 3
RESULTS AND DISCUSSION

3.1. Nitrosamine Formation Catalysis

Previous research from our group (24, 25) showed that secondary amines could be adsorbed onto ACs and combine with reactive oxygen and nitrogen species present on the carbon surfaces to form nitrosamines. These reactions required atmospheric contact with the carbon materials and formation increased significantly with longer air contact time. It was thus necessary to first prove that AC can catalyze formation of nitrosamines without oxygen from the air when in the presence of a nitrosating agent such as nitrite. For this ‘proof of concept’ study, slightly higher concentrations and carbon loadings were used than for all subsequent studies. DMA and NO₂⁻ were spiked at 200 µM and 200 mg AC was added to the 250 mL samples. Five different sample types were prepared: AC only (without NO₂⁻ or DMA), AC and NO₂⁻, DMA and NO₂⁻ (without carbon), AC and DMA, and one set with AC, NO₂⁻ and DMA. Their corresponding NDMA formations were 0.70 nM (± 0.25), 0.75 nM (± 0.49), 3.27 nM (± 0.24), 4.10 nM (± 1.54), and 303.39 nM (± 7.21). The outcome of this experiment supported the hypothesis that carbons can transform amines and nitrite into nitrosamines without the influence of atmospheric contact of nitrogen and oxygen during drying periods. Furthermore, the nitrosamine formation was 74 times greater in the presence of AC than in the absence of AC, suggesting that the presence of AC is required to catalyze nitrosation under the conditions employed to generate appreciable amounts of nitrosamines.
To further investigate the impact of dissolved oxygen on amine nitrosation catalysis, a set of experiments were conducted under limited oxygen conditions by purging solutions with nitrogen gas. Aqueous samples containing 100 mg carbon and 50 µM of DMA and NO$_2^-$ were purged with N$_2$ gas for 30 minutes to remove any significant levels of dissolved oxygen then sealed with Teflon lined caps prior to agitation. Four sample sets were created for this analysis: AC+DMA+NO$_2^-$ purged with N$_2$ gas, AC+DMA+NO$_2^-$ without purging, DMA+NO$_2^-$ purged with N$_2$ gas (no AC), and DMA+NO$_2^-$ without purging (no AC). The resulting NDMA formations were 33.46 nM (± 2.60), 37.39 nM (± 1.05), 1.67 nM (± 0.20), and 1.58 nM (± 0.05), respectively. It can be concluded from these results that dissolved oxygen had negligible impact on the overall NDMA formation for each sample, regardless of the presence of AC. Formation was substantially higher with the AC samples, further supporting the catalytic impact of AC on nitrosation reaction.

3.2. AC Particles versus Cartridges

The most commonly applied method for nitrosamine analysis requires aqueous samples to be passed through AC packed media cartridges for SPE; this method varies slightly from the protocol used in this study. Since we are primarily interested in uncovering the factors that determine the magnitude of nitrosamine formation in wastewater conditions, it is beneficial to use the particle method detailed above to maximize contact between carbon particles and aqueous precursors by allowing AC to suspend freely in conditions similar to a batch reactor. Consequentially, it was important to directly compare the NDMA formation potential between these two methods. Five-hundred mL samples containing equimolar concentrations of DMA and nitrite were processed according to each of the prescribed methods across a range of amine spiking conditions. Due to the higher carbon usage compared to following studies, the analytical procedure employed 10 mL (compared to 1.5 mL in the typical protocol) of DCM to
mobilize the sorbed nitrosamines to ensure complete extraction. Results from these experiments are shown in Figure 1.

**Figure 1.** Particle method vs. cartridge method. ■: NDMA, –: DMA, •: NO₂⁻. pH 7.5, 4 g/L conditioned cartridge carbon, DMA and NO₂⁻ varied from 50-200 µM.

Adsorption trends were similar for the particle and cartridge methods; i.e., the amounts of DMA or NO₂⁻ adsorbed to the AC were comparable. As anticipated, the particle method resulted in higher NDMA formation, although measured values were on the same order of magnitude for both methods. This outcome suggests that results obtained using the particle method are proportional to those gathered from the cartridge method when the precursor concentration was 100 µM or lower, although slightly amplified due to a higher effective contact time since the particle method allows AC to be fully submersed instantaneously. These results suggest that AC catalyzed nitrosation may be more significant than previously thought on analytical methods and even under wastewater treatment conditions.
3.3. Precursor Concentration Ratios

The ability of AC to catalyze nitrosamine formation from amine and nitrite precursors demonstrates the importance of surface reactions on AC. In practice, real water samples contain diverse constituents at various levels which may complicate analysis and lead to measurement errors. Since AC particles have a finite amount of surface sites available for adsorption, select ions may preferentially adsorb and leave others suspended in the aqueous media. Even for just the two precursors alone, higher concentrations of one precursor may allow that compound to outcompete the other dissolved chemical species for adsorption sites and alter the nitrosamine formation potential of the water. To demonstrate this incidence, two sets of samples were prepared in which one precursor was held constant at 50 µM while the other, labeled ‘X’, was varied from 20-200 µM. Results from these experiments are shown below in Figure 2.

![Figure 2](image)

**Figure 2.** Effect of precursor variation. pH 7.5, 400 mg/L AqC carbon, either DMA or NO$_2^-$ varied from 20-200 µM while other analyte held constant at 50 µM.

When DMA was the varied analyte, NDMA formation was generally higher, while the DMA adsorption rate was fairly constant at around 90%. Nitrite was less favorably adsorbed in all samples measured (<30%) and less nitrosamine was formed
with excess NO\textsubscript{2} compared to samples with excess DMA, suggesting that the AC-catalyzed nitrosamine formation potential from amine and nitrite is limited by secondary amine concentration, rather than that of NO\textsubscript{2} under the conditions employed. This finding is also in contrast to the known nitrosation kinetics in aqueous solutions, in which the nitrosation rate has a greater dependence on the nitrosating agent (nitrous acid) concentration (rate = \( k[HNO_2]^2[\text{amine}] \)) (32). The adsorption affinity of inorganic nitrite ion to the organic AC surfaces is expected to be low. The greater dependence of the AC-catalyzed nitrosation on amine concentration is consistent with the stronger interaction of amine with AC surfaces than nitrite.

**3.4. Nitrite versus Nitrate**

Although nitrite was chosen as the focus for our experiments due to its ubiquity in wastewater influents and ease of preparation, gaseous nitrosating agents such as N\textsubscript{2}O\textsubscript{4} and N\textsubscript{2}O\textsubscript{3} have also been shown to convert amines to nitrosamines (20). Moreover, nitrate may be reduced to nitrite and lead to an additional source of precursor that, although studied extensively (33, 34) for relevance as an in-vivo nitrosating agent, has been largely overlooked for its potential to form residual nitrosamines in wastewater effluent streams. To place the scope of this phenomenon into perspective, a set of experiments were conducted to compare the efficacy of nitrate and nitrite in converting DMA into NDMA at neutral pH ranges in the presence of granular activated carbon (AqC) or PAC. The results of this analysis are shown in Figure 3. A similar analysis for NMOR can be seen in Figure 4.
Figure 3. Nitrosating agent comparison. pH 7.5, 800 mg/L AqC carbon, DMA spiked at equimolar concentrations with either nitrate or nitrite.

Figure 4. Morpholine nitrate and nitrite comparison. pH 7.5, 800 mg/L AqC carbon, MOR spiked at equimolar concentrations with either nitrate or nitrite.

DMA or MOR were spiked at equimolar concentrations with each of the nitrosating agents shown above. Powdered carbon formed the greatest amount of NDMA and NMOR, presumably due to its higher surface area available for adsorption and
transformation of the precursors (1,346 m$^2$/g vs. 1,202 m$^2$/g) (25, 35). Both PAC and AqC formed significant levels of nitrosamines, while nitrite proved to be a more efficient nitrosating agent than nitrate. Since NO$_2^-$ or NO$_3^-$ must first be converted to their protonated forms before catalyzing nitrosation, it is easier for nitrite (pKa = 3.4) to become nitrous acid than it is for nitrate (pKa = -1.4) to become nitric acid (32).

### 3.5. Effect of Amine Structure

Recent studies have reported nitrosamine formation due to chloramination of waters polluted with both secondary and tertiary amines (36, 37, 38). Additionally, select tertiary amines have been shown to undergo nitrosation by cleavage of larger functional groups to form DMA which can then be transformed (39). We selected several secondary and tertiary amines that have been shown to result in the most significant nitrosamine formation by chloramination as representatives for further analysis of potential AC-catalyzed nitrosation. The secondary amines included DMA, PYR, DEA, MOR and DBA. Figure 5 shows the structures of the selected tertiary amines.

![Tertiary amine structures](image.png)

**Figure 5.** Tertiary amine structures.

Figure 6 shows the resulting nitrosamine formation for each of the tested amines. All secondary amines form their corresponding nitrosamines by replacing their hydrogen
atoms with a nitroso group. DBA decidedly formed the most of its respective nitrosamine (i.e., NDBA), which was consistent with expectations based on previous experimental results (25). With the exception of DMA, secondary amines were transformed in the order of increasing molecular weight (MW), although there was no clear correlation with respect to amine adsorption. Figure 7 reaction 1 shows the reaction pathway for nitrosation of secondary amines to yield nitrosamines. The kinetics of direct nitrosation is second order for nitrite and first order for amines (20). The higher transformation of MOR to NMOR and DBA to NDBA may be due to: (1) the relatively more rapid nitrosation of MOR and DBA ($k_{MOR} = 6.7 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$, $k_{DBA} = 5.4 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$) compared to other secondary amines ($k_{DMA} = 3.6 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$, $k_{DEA} = 3.3 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$), where nitrosation rate = $k_{AMINE}[\text{HNO}_2]^2[\text{amine}]$ (32), and (2) the strong adsorption of MOR and DBA to AC surfaces due to their larger MW and higher hydrophobicity.

![Figure 6](image-url)

**Figure 6.** Nitrosamine formation for various amines. pH 7.5, 400 mg/L AqC carbon, NO$_2^-$ and amine spiked at 50 µM.
Secondary amines:

\[
R_2NH + HNO_2 \rightarrow R_2NNO + H_2O \quad (1)
\]

Tertiary amines:

\[
R_2NCHR_2' + HNO_2 \rightarrow R_2N^+CHR_2' + NO \quad (2)
\]

\[
R_2N^+CHR_2' + H_2O \rightarrow R_2C=O + R_2N^+H_2 + HNO_2 \rightarrow R_2NNO \quad (3)
\]

\[
2HNO \rightarrow H_2N_2O_2 \rightarrow H_2O + N_2O \quad (4)
\]

**Figure 7.** Nitrosamine formation from nitrosation of secondary and tertiary amines (39).

While the secondary amines formed their corresponding nitrosamines, all the selected tertiary amines formed NDMA (Figure 6). Although TMA formed relatively low levels of NDMA, the remaining three tertiary amines were converted to NDMA with comparable efficiency to the conversion of lower MW secondary amines to their corresponding nitrosamines. Figure 7 reactions (2-4) show the proposed pathways for nitrosation of tertiary amines to yield nitrosamines (39). According to Smith and Loeppky (39), tertiary amines may react with aqueous nitrous acid to form an N-nitrosoammonium ion and *cis* elimination of nitroxyl to form a ternary immonium ion, \( R_2N^+CR_2' \) (reaction 2), which then hydrolyzes to yield a secondary amine that can undergo further nitrosation to yield nitrosamine (reaction 3).

As Figure 5 shows, each of the selected tertiary amines, with the exception of TMA, has an alpha carbon attached to a tetrasubstituted (i.e., quaternary) carbon within an aromatic ring structure. This property theoretically facilitates cleavage of the -N(CH$_3$)$_2$ group due to the relative stability of the leaving groups, resulting in a greater potential for nitrosamine formation. Results show that tertiary amines such as TMA without an alpha
carbon attached to an aromatic structure are less likely to decompose into DMA due to their higher C-N bond strength, and thus should form less NDMA. This hypothesis is consistent with experimental results and provides an accurate means for assessing the likelihood of tertiary amine transformation based on structural properties.

3.6. Effect of Reaction Conditions

3.6.1. Kinetics

Since wastewater treatment facilities operate under a wide range of conditions, it is important to place the role of environmental and reaction conditions into context. A kinetics study, whose results are shown in Figure 8, revealed that precursor adsorption peaks relatively early for both DMA and NO$_2^-$, with equilibrium conditions reached at around 8 hours. Despite this outcome, NDMA formation continued to increase until particle contact time reached 4 days. It can thus be concluded that nitrosamine formation is governed by the transformation reaction, rather than adsorption rates.

![Figure 8. Reaction and adsorption kinetics. pH 7.5, 400 mg/L AqC carbon, 50 µM DMA and NO$_2^-$](image)
3.6.2. pH

While the pH trend of nitrosation has been clearly demonstrated to result in higher formation in more acidic environments, there has been only one study to our knowledge that addressed the relevance of pH to AC catalyzed nitrosation (23). Dietrich et al. concluded that lower pH values enhance nitrosation by granular activated carbon particles in agreement with previous hypotheses. Their study, however, utilized only one type of amine (N-methylaniline) and two pH values (pH 4 and 7). Samples prepared at pH 4 resulted in N-nitroso-n-methylaniline (NNMA) formation approximately 2 orders of magnitude higher than at pH 7. For our purposes, a systematic pH study was conducted by spiking 10 mM of phosphate buffers prepared at the specified pH ranges into solutions containing 50 µM nitrite and 50 µM of the secondary amines shown in Figure 9, and final solution pH values were verified by measurement after mixing.

![Figure 9. Results from pH study. 400 mg/L AqC carbon, 50 µM amine and NO₂⁻.](image)

In conjunction with the previously published pH trend data, the results of this study show that the optimum nitrosation pH is highly dependent on the amine being transformed. Furthermore, nitrosamine formation trends peaked near the pKa values for each of the amines (DMA = 10.6, MOR = 8.4, NMA = 4.9) which indicates that speciation plays a large part in transformation. While Dietrich et al. reported much higher...
formation of 524 µM NNMA from 933 µM spiked NMA (approximately 56% conversion at their highest carbon loading and nitrite concentration), the absolute values are less important to compare than the overall trends since several reaction conditions were different between the two studies. It is also important to note that this pH study utilized only one type of carbon with a pH$_{PZC}$ measured at 9.6, while different materials may further complicate the reaction dynamics.

3.6.3. Phosphate Buffer

Phosphorus is present in a wide variety of wastewaters and may alter the dynamics of nitrosamine formation. Analytical methods also require the use of buffer solutions to prevent large variances and inaccurate measurements. Xu et al. reported the role of phosphate in diminishing NDMA formation from nitrosation of DMA in aqueous mixtures and hypothesized that the cause was due to complexation of sodium nitrite with monosodium phosphate present in the solution (40). However, this potential interference by phosphate has not yet been investigated with respect to AC-catalyzed nitrosation reactions and at realistic precursor concentrations. To assess the effect of phosphate buffers on nitrosamine production in the presence of AC, samples were prepared containing 50 µM of each DMA and NO$_2^-$, 400 mg/L of AC, and phosphate buffers ranging from 5-25 mM while using NaCl to maintain overall ionic strength consistent at 0.1 M. Figure 10 shows evidence that confirms the findings of the aforementioned study at environmentally relevant concentrations, i.e., increasing phosphate concentration hindered the AC-catalyzed nitrosation.
Figure 10. Phosphate inhibition study. pH 7.5, 400 mg/L AqC carbon, 0.1 M ionic strength, 50 µM DMA and NO$_2^-$.

The adsorption trends shed new light on the impact of phosphate since nitrite adsorption increased with increasing phosphate concentration. If, in fact, complexation occurred between NaH$_2$PO$_4$ and NaNO$_2$ as suggested by Xu et al. (40), it is likely that these complexes become more prone to take up space on AC particles and prevent DMA from adsorbing. Additionally, the relatively smaller amount of DMA adsorbed in the higher phosphate concentration samples limited the potential for NDMA formation. It is important, therefore, to ensure that the same type and concentration of buffer was used when comparing nitrosamine measurements from different laboratories.

3.7. Effect of Carbon Modification

It is thought that the surface functional groups and general AC properties should considerably alter NDMA formation by creating more or less reactive sites for transformation to occur. Previous study from our research group (25) has outlined the effects of various treatments on carbon surfaces: acid treatment oxidizes particles and decreases pH$_{PZC}$, whereas heating the particles releases entrapped oxygen and basifies the surfaces. *Tert*-butyl alcohol (TBA) may be used to quench radical oxygen species in aqueous solutions, which was expected to reduce nitrosamine formation since these
compounds have been hypothesized to influence AC catalyzed amine transformation. Figure 1 shows the outcome of various carbon treatments.

![Graph showing the effect of different carbon treatments on NDMA formation. The x-axis represents the type of carbon treatment, and the y-axis represents NDMA formed (nM). The treatments include TBA, 600°C, H2SO4, NanoAg, and HNO3. NDMA formed increases significantly with NanoAg and HNO3 compared to other treatments.]

**Figure 11.** Effect of carbon treatment. pH 7.5, 400 mg/L AqC carbon, 50 µM DMA and NO2−.

TBA treatment reduced NDMA formation by approximately 22% as anticipated, but heat treatment and sulfuric acid treatment had little effect on experimental results. Impregnation of AC particles with approximately 3.27 mg/g nanosilver particles (NanoAg) resulted in a spike in NDMA formed, although there was no clear trend with respect to precursor adsorption for any of the modified carbons tested. This result is concerning since several commercially available products utilize silver nanoparticle doped AC to prevent bacteria from accumulating on particle surfaces; such amended particles may result in significant nitrosamine formation when filtering waters impacted by nitrosating agents. Soaking particles in concentrated nitric acid solution resulted in a surge in NDMA formed, despite the negligible nitrite adsorption. It is therefore likely that this change is attributable to residual nitrosating agent within the AC pores.
3.8. Environmental Relevance of Findings

To determine the effect of real water matrices, samples were prepared using the protocols outlined in the ‘AC Particles versus Cartridges’ study and total precursor concentrations (background + spiked) of 50 µM for DMA and NO$_2^-$\. Wastewater was collected from a secondary effluent sampling point (after activated sludge treatment and clarification and before chlorination) and had an initial pH of 6.83, alkalinity of 119 mg/L as CaCO$_3$, and ammonia, phosphorus, nitrate, nitrite, and DMA concentrations of 12.35 µM, 1.47 µM, 46.77 µM, 0.97 µM, and 0.07 µM, respectively. Nitrosamine formation for both methods was approximately 62% higher in the DI water samples than in the real wastewater. This result is likely due to interaction of precursors with other dissolved constituents present in wastewater. It is also possible that other organic constituents in the wastewater competed for AC surface sites and resulted in less precursor adsorption and thus less NDMA formation. Amine adsorption data confirmed this hypothesis since approximately 7.6% more DMA was adsorbed in DI water than in wastewater. Despite the diminished formation in wastewater, NDMA was still formed at 17.96 ± 0.08 nM, approximately 80% higher than the notification level in California (41) and 26 times of the $10^{-6}$ cancer risk concentration (1).
CHAPTER 4

CONCLUSIONS

The results presented in this paper highlight the importance of a previously understated phenomenon. Activated carbon has been shown to catalyze the transformation of both secondary and tertiary amines under ambient conditions. While previous studies have claimed that nitrosation occurs ideally under acidic conditions, our data shows that maximum formation occurs near the pKa value for each individual amine when in the presence of AC materials. As a direct consequence of this outcome, amines with acid dissociation constants within the neutral pH range that wastewater treatment facilities typically operate may be isolated to effectively meet future regulatory requirements. Since nitrite has been shown to be a relatively effective nitrosating agent, it may be beneficial to further remove nitrite to below the current effluent requirements to minimize nitrosamine contamination. While this study has laid the groundwork by systematically addressing the contribution of individual precursors toward nitrosamine formation, it is important that future studies address the combined effects of multiple precursor streams since contaminants are rarely isolated in complex natural or engineered systems.
APPENDIX A

GC-MS CONDITIONS

The conditions used for nitrosamine and amine analysis mimic those used in our previous related studies. An Agilent GC-MS (6890/5973) system, equipped with a HP-5MS column (30 m x 250 μm x 0.25 μm), was used to process and analyze samples. A large volume injector was used for nitrosamine analysis, per EPA Method 521, and solvent-vent mode injections of 5-25 μL were made for secondary amines and nitrosamines, respectively. The initial injection port temperature was 45 °C, and was ramped to 210 °C at a rate of 720 °C per minute (held for 4 minutes), followed by a second increase to 250 °C at 500 °C/min (held for 5 minutes). The oven temperature program for nitrosamine samples was as follows: 45 °C held for 1 minute, then ramped to 70 °C at 10 °C/min, followed by another increase to 220 °C at 18 °C/min. For secondary amines, the oven settings were: 45 °C held for 1 minute, followed by an increase to 260 °C at a rate of 20 °C/min (held for 5 minutes). Electron ionization mode was used in conjunction with selected ion monitoring mode. Parent and daughter ions are shown in Table A for each analyzed species along with their respective retention times using this method.
Table A. GC-MS Monitoring Ions and Retention Times.

<table>
<thead>
<tr>
<th>Type</th>
<th>Species</th>
<th>MW (g/mol)</th>
<th>Monitoring Ion (m/z)</th>
<th>Retention Time (min)</th>
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<td>Nitrosamine</td>
<td>NDMA-d6</td>
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<td>80.1</td>
<td>6.0</td>
</tr>
<tr>
<td></td>
<td>NDMA</td>
<td>74.1</td>
<td>74.1</td>
<td>6.0</td>
</tr>
<tr>
<td></td>
<td>NMOR</td>
<td>116.1</td>
<td>116.1</td>
<td>9.1</td>
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<tr>
<td></td>
<td>NDBA</td>
<td>158.2</td>
<td>84.1</td>
<td>10.4</td>
</tr>
<tr>
<td></td>
<td>NDEA</td>
<td>102.1</td>
<td>102.1</td>
<td>7.1</td>
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<tr>
<td></td>
<td>NPYR</td>
<td>100.1</td>
<td>100.1</td>
<td>9.3</td>
</tr>
<tr>
<td>Secondary Amine</td>
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<td>191.3</td>
<td>11.4</td>
</tr>
<tr>
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<td>DMA</td>
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</table>
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