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Micro-structured copper and nickel metal foams for wastewater disinfection: proof-of-concept and scale-up

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It is necessary to disinfect treated wastewater prior to discharge to reduce exposure risks to humans and the environment. The currently practiced wastewater disinfection technologies are challenged by toxic by-products, chemicals and energy demand, a range of effectiveness limitations, among other concerns. An effective, eco-friendly, and energy-efficient alternative disinfection technique is desirable to modernize and enhance wastewater treatment operations. Copper and nickel micro-structured metal foams, and a conventional copper mesh, were evaluated as disinfecting surfaces for treating secondary-treated wastewater contaminated with coliform bacteria. The micro-structured copper foam was adopted for scale-up study, due to its stable and satisfactory bactericidal performance obtained over a wide range of bacterial concentrations and metal-to-liquid ratios. Three scales of experiments, using two types of reactor designs, were performed using municipal wastewater to determine the optimal scale-up factors: small lab-scale batch reactor, intermediate lab-scale batch reactor, and pilot-scale continuous tubular reactor experiments. The performance was evaluated with the aim of minimizing metal material requirement with respect to bactericidal efficiency and leaching risks at all scales. Copper foam, at or above optimal conditions, consistently inactivated over 95% of total coliforms, fecal coliforms and E.coli in wastewater at various scales, and leachate copper concentrations were determined to be below Canadian guideline values for outfall. This study successfully implemented the “structure” strategy of process intensification, and opens up the possibility to apply micro-structured copper foam in a range of other water disinfection systems, from pre-treatment to point-of-use, and should thus become a topic of further research.

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1. Introduction

Disinfection is one of the indispensable processes to inactivate disease-causing microorganisms in wastewater treatment plants (WWTPs) effluent prior to outfall. The most widely used disinfection technologies are chlorination and ultraviolet (UV) irradiation (Leong et al., 2008). However, the challenges faced with these two techniques are raising concerns, such as the disinfection by-products (DBPs) generation after chlorination, and the energy-consumption and influent turbidity limitations associated with the performance of UV irradiation (Du et al., 2017; Liu and Zhang, 2006). There are several other disinfection technologies available that are less commonly applied in WWTPs, such as ozonation and reverse osmosis. The limited number of applications are due to high capital and/or operating costs, ineffective performance for specific pathogens, or by-products concerns (Chidambaranathan and Balasubramaniam, 2019). Accordingly, there is still an essential need for an alternative disinfection technology that can achieve high disinfection performance while being eco-friendly, energy-saving, simple to run, and economically viable.

Microbial contact with metal surfaces is considered to be an alternative form of disinfection technology in sanitary applications. The U.S. Environmental Protection Agency has recognized copper as a solid antimicrobial material that could be applied in facilities with a high sanitary requirement such as hospitals (Colin et al., 2020, 2018; Molteni et al., 2010). Most recently, copper surfaces have even been shown to be capable of combating the spread of the coronavirus, including COVID-19 (van Doremalen et al., 2020; Warnes et al., 2015). Bright et al. (2009) studied the anti-viral effect of zeolites loaded with metal ions, and tested copper due to its toxic to many microorganisms as a result of its ability to block functional groups on proteins and inactive enzymes, thus preventing both enveloped and unenveloped viruses from being able to enter human cells. Such zeolite powders could be added or bonded to materials such as plastics, paints, synthetic fabrics, and steel surfaces. For water and wastewater applications, there are two forms

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of metal materials that have been considered for disinfection: metal nanoparticles and metal surfaces (Chakraborty et al., 2017; Cui et al., 2012).

The antimicrobial properties of metal nanoparticles have been widely studied and enunciated in the applications of water disinfection as an alternative water and wastewater treatment solution (Cui et al., 2012; Dankovich and Smith, 2014; Lv et al., 2018). Metal nanoparticles, of appropriate antimicrobial composition, have been demonstrated to be highly effective on microorganism inactivation due to their high specific surface area. Lv et al. (2018) reported that the antibacterial efficiency of copper nanoparticles could reach 94.3 ± 0.1 % after a 12-h treatment cycle with an initial Escherichia coli (E. coli) concentration at 10⁵ colony-forming unit (CFU)/mL. In another study, silver nanoparticles were embedded in a blotter paper to inactivate bacteria while water percolated through (Dankovich and Gray, 2011). The result supported the disinfection potential of metal nanoparticles since a log 7.6 ± 1.3 reduction of E. coli was achieved in 10 min percolation compared to the 10⁶ CFU/mL influent concentration. However, the complex and costly synthesis processes, tendency to easily aggregate, and low reusability of nanoparticles have limited their applicability in large-scale applications (Dizaj et al., 2014; Musarrat et al., 2010; Villaseñor and Rios, 2018). The potential impacts of metal nanoparticles in the environment and on human health is another concern for its application in water and wastewater treatment (Kim et al., 2013; Scown et al., 2010).

The use of metal surfaces as bactericides has its origin in the search for materials to be incorporated into object surfaces to prevent microbes. A typical setting for this application is hospital wards (Michels et al., 2009), food processing facilities (Wills et al., 2005) and other public areas (e.g. public transport) where people are susceptible to topically contacting pathogens. The main antimicrobial mechanisms are attributed to cell damage when bacteria come into contact with metal surfaces, leading to rapid cell death, are cell membrane rupture, protein disruption and Fenton-type reaction. For water treatment application, Varkey and Dlamini (2012) used copper mesh inside clay and sawdust filtration pots, meant for small-scale applications, and observed complete elimination of E. coli bacteria. Varkey (2010) also tested thin metal plates immersed in contaminated water for E. coli inactivation, and found that copper, silver and zinc had higher rates of destroying the bacteria than aluminium, tin and silicon, and that greater surface area led to faster coliform elimination. However, the studies on the application of metal surfaces are limited and no experimental data are available on either wastewater treatment or larger scale experiments beyond laboratory experimentation, which is insufficient to demonstrate the wastewater disinfection potential of metal surfaces.

The present study was carried out to determine the practicability of micro-structured metal foams, similar to those used in hybrid-car battery technology, to disinfect wastewater treatment effluent, as a process intensification (PI) approach. The use of micro-structure foams for wastewater disinfection constitutes an application of the “structure” domain of PI, with the objective to “maximize the effectiveness of molecular events” (Santos and Van Gerven, 2011); in wastewater disinfection the molecules are the pathogens, and the events become the pathogen-metal contact. Such innovative water disinfection technology could supplant or work in conjunction with commonly used sterilization processes found in wastewater or drinking water treatment plants. Metal foams have high (≥95 %) porosity, which minimizes pressure losses as water travels through it, while having high surface area for effective contact between the metallic surfaces and the flowing water. Such foams are also metallurgically stable and resist dissolution in water. These properties should allow the development of a continuous flow-through bed treatment system that can be compact and long-lasting, while having little impact on water flow or composition. The main purpose of this study was to examine the bactericidal efficiency of metal foams at lab- and pilot-scales fed with municipal WWTP secondary-treatment effluent. A scale-up study was conducted to investigate the efficiency of the metal foams while treating a large volume of water over extended periods of time, and the scale-up factors were determined for further technology development. Metal leaching risk was also assessed to evaluate the potential impacts on environmental and human health.

2. Materials and methods

2.1. Metal materials

The nickel foam and copper foam materials were supplied by Cnem Corporation (Mississauga, Ontario, Canada). The density, porosity and pore density of copper foam were, respectively, 215 kg/m³, 95 % and 110 ppi (pores per linear inch). For the nickel foam, these values were 292 kg/m³, 95 %, and 90 ppi. Fig. 1 illustrates the micro-structure of the metal foams observed using a digital microscope (VHX-5000, Keyence). The metal struts form a mesh structure with fully open and interconnected pores. For comparison, a common copper mesh was also used, supplied by Bird B Gone (Irvine, California, USA) for household usage, and the density, porosity, and pore density the copper mesh were determined to be 197 kg/m³, 90 %, and 4.6 ppi.

2.2. Bactericidal testing

E. coli CMF-Sh1 strain (ATCC® 43,651™) was purchased from ATCC (Manassas, Virginia, USA). The E. coli strain was activated by incubating it into autoclaved agar medium and incubating it under 37 ± 2 °C for 24 h following the “ATCC® bacterial culture guide” (ATCC, 2015). The typical E. coli concentrations found in treated wastewater prior to disinfection in published literature were reviewed and classified into three levels: low concentration (Boutilier et al., 2009; Wen et al., 2004), moderate concentration (Lazarova et al., 1999; Pérez et al., 2010), and high concentration (Le-Thi et al., 2017; Wang et al., 2007). The dosage of the bacterial stock solution was adjusted in order to achieve low concentration (10^2-10^3 CFU/mL), moderate concentration (10^3-10^4 CFU/mL) and high concentration (10^4-10^5 CFU/mL) in autoclaved water. Water used to prepare the bacterial solutions was previously treated by Milli-Q® IQ 7003/05/10/15 water purification system. The bacterial solutions thus prepared were used for bactericidal performance comparison of metal materials in lab-scale testing. Killing rate (KR) was determined by Eqn. (1):

\[
KR(\%) = \frac{C_0 - C_1}{C_0} \times 100\%
\]

where \(C_0\) (CFU/mL) and \(C_1\) (CFU/mL) are the bacterial counts of solutions before and after metal exposure, respectively.

2.3. Reactor design and scale-up testing

Three reactor scales (small lab-scale, intermediate lab-scale, and pilot-scale) were used for testing the scale-up potential of the conceptualized process. Bactericidal efficiency, retention time and metal material mass were considered to evaluate performance.

The small lab-scale reactor was designed, as demonstrated in Fig. 2a, to model a continuous stirred tank reactor (CSTR). Copper foam, copper mesh, and nickel foam were used to construct reactors. The metal materials were shaped into three layers of cylinders with increasing diameters and stabilized by thin copper wire to maintain an equal gap between the concentric cylinders. The metal materials were placed within beakers of two sizes (600 mL,
Fig. 1. (a) Image of the copper foam structure; (b) Image of the nickel foam structure.

Fig. 2. (a) The small lab-scale reactor setup: a-1: Air bubbling inlet; a-2: Supporting wire; a-3: Metal cylinders; (b) The intermediate lab-scale reactor setup: b-1: Air bubbling inlet; b-2: Metal cylinders; (c) The pilot-scale reactor setup: c-1: Inflow; c-2: Valve 1; c-3: Valve 2; c-4: Valve 3; c-5: Valve 4; c-6: Outflow.
and 2000 mL) containing between 300 mL and 2000 mL of bacterial solution or of real secondary-treatment effluent from the Guelph WWTP. The metal mass of the three tested materials was kept constant at 5.5 g for all tests. Air pumps (Tetra 77.847) were applied to supply air bubbling throughout the experiment in each reactor to ensure aeration and agitation. The experiments were conducted under room temperature. Control group (C), copper foam group (CF), copper mesh group (CM), and nickel foam group (NF) were set in the beakers with no material, copper foam material, copper mesh material, and nickel foam material, respectively. Treated solution samples were collected after 0.5 h, 1 h, 2 h, 3 h and 4 h of contact time. Bactericidal tests were performed in triplicate for comparing the bactericidal performance of different metal materials, and selecting the suitable metal material for scale-up study.

The intermediate lab-scale reactor enlarged the lab-scale reactor for scale-up study, being itself also a CSTR-style reactor equipped with concentric metal foam cylinders, as illustrated in Fig. 2b. Three air pumps were applied in the intermediate CSTR to provide effective mixing. The reactor capacity was enlarged to 18.9 L, within which 9.0–10.0 L of real secondary-treatment effluent from the Guelph WWTP was treated by 85 g–183 g of copper foam; varying liquid volume and metal mass allow to study the effect of metal-to-liquid ratio on bactericidal performance. Wastewater samples were collected from the secondary treatment effluent on the day of each experiment. The secondary effluent had passed through a pump station, screen, grit chamber, primary clarifier, aeration tank, and secondary clarifier, before collecting. The secondary wastewater contained an average concentration of total coliform, fecal coliform, and E. coli of 621 ± 173 CFU/mL, 82 ± 28 CFU/mL, and 111 ± 23 CFU/mL, respectively. Wastewater samples were stored in a cooler at a temperature of 1–4°C during transition to the laboratory. In the intermediate lab-scale testing, treated wastewater samples were collected after 0.5 h, 1 h, 2 h, 3 h and 4 h of contact time.

To achieve the goals of space-saving and higher efficiency, the plug flow reactor model, under continuous operation, was applied in the pilot-scale experiment for further scale-up study. Plug flow reactors are ideal for continuous processing and ensure tight residence time distribution in a reduced reactor volume compared to CSTR design. The pilot-scale setup is illustrated in Fig. 2c as was operated at the Guelph WWTP piloting facility, which continuously receives wastewater streams for testing directly from the full-scale municipal plant. The length of straight PVC pipes was 6.16 m in total, and connected by six 4-inch-diameter PVC elbows to create three horizontal legs stacked over each other. The reactor was fed with secondary treatment effluent (sampled for each experiment, and analyzed for total coliform, fecal coliform, and E. coli) from an overhead tank continuously, and a constant fluid height was maintained at the inlet vertical leg to ensure constant pressure head to drive the flow at a constant flow rate. The inflow rate was measured at 1.207 ± 0.04 L/min. The flow outlet was located above the height of the highest horizontal leg, and the horizontal legs where stacked at different heights, to ensure equal residence time for all flow exiting the reactor. The total liquid volume at any given time within the pilot-scale reactor was 58 L, resulting in a total residence time (from inflow to outflow) of 28.0 ± 0.5 min. Four valves were installed along the length of the reactor for periodically collecting treated wastewater samples. Circular-cut portions of copper foam sheets (selected based on its performance in lab-scale tests) were stacked into cylinder-shaped stacks that snugly fit inside the PVC pipes to evenly occupy the cross-sectional flow area, and the wastewater percolated through eight foam sections (145 g each, 1160 g total) before reaching the outflow of the reactor. Each experimental run began with the reactor empty, and wastewater being fed to the inlet. The treated wastewater samples were collected from four valves located along the length of the reactor, at five points in time: when the wastewater first reached a given valve, and then 0.5 h, 1 h, 2 h, 3 h and 4 h after that. In this manner, the fluid volume from which each sample was taken at a given timestamp would correspond to the same fluid volume from which samples with the same timestamp were taken at each valve. The difference between such samples from the same fluid volume being how many copper foam sections the fluid volume travelled through (metal contact area) and how much time the fluid volume spent within the reactor (residence time).

2.4. Water analyses

The membrane filtration technique was adopted from the procedure outlined in the EPA Methods 9132, 1603 and 1604 (U.S. EPA, 2002a, 2002b, 1986) for trapping total coliform, fecal coliform, and E. coli. The subsequent enumeration was conducted by cultivation on different mediums. Millipore membrane filters with 0.45 μm pore size, Membrane Endo Agar (M-Endo), Membrane Fecal Coliform Agar (MFC), and Plate Count Agar (PCA) were acquired from Fisher Scientific (Waltham, Massachusetts, USA). Plates were prepared for enumerating total coliforms, fecal coliforms, and E. coli. The filters with trapped bacteria were placed on Petri dishes containing a layer of the respective mediums. The inoculated Petri dishes were then separately incubated at 37°C and 44.5°C for 12 h to generate countable plaques. The detection limit for wastewater samples was 10 CFU/mL for total coliforms and 2 CFU/mL for fecal coliforms and E. coli. KR was determined by Eqn. (1). All the microbial counting readings were taken in triplicate, and the means are reported in this study. Except for optimization experiments, all other data were expressed as mean ± standard deviation (SD) based on the execution of three independent experiments.

Water samples were also monitored for pH, and dissolved oxygen (DO), in triplicates. The statistical difference of monitoring parameters and KR values between various treatment groups in the lab-scale experiments was assessed by one-way ANOVA. P < 0.05 was used to determine the significant level.

2.5. Metal leaching

The loss of metal ions to the treated wastewater after metal material contact was assessed by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) NexION 2000 from PerkinElmer (Waltham, Massachusetts, USA). Water samples were collected and autoclaved after four-hour treatment cycle by metal materials. Autoclaved samples were filtrated by 0.2 μm membrane filter (Fisher Scientific, 13,100,112), and diluted with 1 vol% nitric acid solution containing Indium as internal standard, then analyzed by ICP-MS for Cu and Ni ionic concentration. Standard solution calibration curves had a correlation coefficient of over 0.995. The analyses were conducted in triplicate, and the detection limit was 0.1 μg/L.

3. Results and discussion

3.1. Comparison of metal materials

The KR of different treatment groups were determined under three different levels of bacterial concentration in Milli-Q water solutions (low concentration, moderate concentration, and high concentration), using the small lab-scale CSTR-style reactor and the prepared bacterial solutions. The bactericidal results are presented in Fig. 3a, b, and c, respectively.

The initial bacterial count of the low concentration solution was at 202 ± 15 CFU/mL. The control group had a decreasing bacterial concentration with time due to these bacterial solutions being prepared by heavily diluting bacterial stock solution using autoclaved Milli-Q water. Thus, all bacterial solutions contained
Fig. 3. (a) Killing rates of different treatment groups under low bacterial concentration; (b) Killing rates of different treatment groups under moderate bacterial concentration; (c) Killing rates of different treatment groups under high bacterial concentration.

minimal amount of nutrients to support the initial bacterial population. What is noticed, however, it that the death rate was drastically sped up by the metal materials. All the mental materials could achieve a 99.0% or greater KR at 0.5 h of retention time. The effectiveness of the metal materials is further supported by the small SD values for those samples, versus the control, which shows that copper and nickel metal contact invariably kills *E. coli*.

For the moderate bacterial concentration experiments, the average bacterial concentration was 3600 ± 361 CFU/mL. Here, as illustrated in Fig. 3b, the control group bacterial concentration remained relatively stable over time, ranging from 3.2 × 10^3 to 3.7 × 10^3 CFU/mL. This suggests that the nutrient content in the moderately diluted stock solution was sufficient to sustain the bacterial population for the duration of the experiment. Under this set of experiments, the disinfection effects of different metal materials slightly differed at 0.5 h of retention time. The average KR after 0.5 h for nickel foam, copper mesh and copper foam were, respectively, 77.7%, 98.5%, and 100%. Again, when KR values were high, the SD values were small, which confirms the consistent performance of the most effective materials. Hence, copper metals, which reached 100% KR in both cases after 1 h retention time, proved to be significantly more effective at rapidly and consistently disinfecting the solution compared to nickel foam.

The high concentration experiment had an average bacterial concentration of 1.6 × 10^5 ± 2.5 × 10^4 CFU/mL. On Fig. 3c, the control group maintained the bacterial concentration during the experiment within a range of 1.3 × 10^5 to 1.7 × 10^5 CFU/mL. From Fig. 3c, it is clear that the copper foam group had a higher KR in short retention time among the three materials, and maintained a 100% KR of *E. coli* throughout the experiment. At 0.5 h, the copper mesh and the nickel foam both had insufficient KR below 75%; however, the KR of copper mesh was able to gradually improve and reach 99.4% after 2 h. Comparatively, the highest KR of nickel foam was 95.6% at 4 h. In conclusion, from these small lab-scale experiments, the results indicate that copper foam is the most effective disinfec-tant material among those tests, with the highest and fastest KR in all three tested solutions.

Considering the economic feasibility, copper and nickel were selected in this study among other metals with antibacterial properties, such as silver and gold. The antibacterial properties of metal copper have gained momentum since 2008, when the U.S EPA registered over 350 copper alloys as antimicrobial (Wei et al., 2014). Comparatively, there are only several studies available on investigating the disinfection efficiency of nickel; however, it has been principally used to facilitate the effectiveness of materials on disinfection, especially those with photocatalytic properties (Applerot et al., 2012; Kruk et al., 2015). From the small lab-scale results, it is clear that three metal materials behave differently with respect to bactericidal performance, depending on retention time and bacterial load. Copper foam and copper mesh exhibited higher bactericidal efficiency compared to nickel foam. In turn, copper foam was substantially more effective under high bacterial concentration due to its micro-structured pores, resulting in greater pore density, and thus increased surface area for contact between the metal, the flowing water, and the suspended pathogens.

The mechanism underlying the antibacterial activities of copper and nickel has been a subject of continued investigation. Some studies indicate that the antibacterial activities of nickel and copper are attributable to metal-induced reactive oxygen species (ROS) generated through Fenton-like reactions (Eqn 2–5) (Applerot et al.,
2012; Grass et al., 2011; Wan et al., 2017). ROS is part of the oxygen metabolism during bacterial growth (Brynildsen et al., 2013), and under natural circumstances cells can detoxify ROS spontaneously through enzymes (Hanukoglu, 2006). Metal materials are postulated to accelerate the generation of ROS, and the excess ROS leads to oxidative damage of cell membranes and DNA, which eventually kill the microorganisms (Wan et al., 2017). The DO level in the small-scale experiments was kept at saturation by aeration for all treatment groups throughout the experiments, and no significant difference (P = 0.98) was observed; thus, sufficient oxygen was available for Fenton reactions to proceed. However, sufficient reduced iron is needed to catalyze the production of hydroxyl radicals, which was not present in these experiments in any detectable amount (i.e. below ICP-MS detection limit).

Nickel-induced Fenton-like reaction (Wan et al., 2017):

\[ Ni + 20_2 \rightarrow Ni^{2+} + 2 O_2^- \quad (2) \]

\[ 2 O_2^- + 2H^+ \rightarrow H_2O_2 + O_2 \quad (3) \]

\[ Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + OH^- + OH^- \quad (4) \]

Copper-induced Fenton-like reaction (Grass et al., 2011):

\[ Cu^+ + H_2O_2 \rightarrow Cu^{2+} + OH^- + OH^- \quad (5) \]

Another hypothesis of the metal-induced disinfection mechanism in wastewater is that bacterial activity on metal surfaces releases metal ions to the solution. The liberated metal ions change the redox potential, neutralize negative charges of the bacterial cell membrane and further disrupt the membrane, which eventually leads to lipid oxidation, DNA lesion, and protein denaturation (Cioffi et al., 2005; Gabrielyan et al., 2016; Hong et al., 2012). The micro-structured copper foam, having greater contact area with the solution, would potentially have greater potential for copper leaching, but only assuming that solubility equilibrium is not reached within the residence time. More on this is discussed in the leaching results, Section 3.4. It is noted, however, that Sicairos-Ruelas et al. (2019) tested the efficacy of 100 μg/L Ag⁺ and 400 μg/L Cu²⁺, individually and combined, as disinfectants against Salmonella enterica serovar Typhimurium, E. coli, Listeria monocytogenes, and Mycobacterium fortuitum, and found that copper did not significantly reduce S. Typhimurium and E. coli on its own, while 2.80-log₁₀ reductions were observed for the other two species.

3.2. Effect of metal-to-liquid ratio

Based on the metal comparison trial results, the copper foam was applied for an optimization study that aimed to minimize the material used to disinfect real wastewater in an acceptable time period. The experiments were conducted with 5.5 g of copper foam to treat adjustable volumes of wastewater, which includes 300 mL, 550 mL, 600 mL, 750 mL, 850 mL, 950 mL, 1500 mL, and 2000 mL. Therefore, the treatment ratios (the mass of metal material to the volume of wastewater) were 1.80 g/100 mL, 1.00 g/100 mL, 0.92 g/100 mL, 0.85 g/100 mL, 0.73 g/100 mL, 0.65 g/100 mL, 0.58 g/100 mL, 0.55 g/100 mL, 0.37 g/100 mL, and 0.28 g/100 mL, respectively. Total coliform, fecal coliform, and E. coli were quantified for evaluating the bactericidal performance. E. coli is recognized as an indicator of fecal contamination in Canada and regulated in recreational guidelines (Tobin and Ward, 1984). Therefore, E. coli was the main parameter in this study for determining the optimal treatment ratio. The other two bacterial counts were used for comparing and supporting the conclusions. The KR values of each group of bacteria, under different treatment ratios, are compared in Fig. 4a, b, and c.

Retention time and KR were considered to be the decisive factors for metal-to-liquid ratio optimization. For meeting the Canadian recreational water quality guidelines, a KR at 96 % is the minimum requirement based on the average E. coli concentration at the Guelph WWTP. The KR as a function of retention time of the three different bacteria groups under the same treatment ratios had slight differences; however, these differences are within an expected variability range for bacterial analysis (within ± 5% differences), except for the lowest ratio. At 0.28 g/100 mL ratio, the three different bacteria groups showed a significant difference in KR, likely due to insufficient contact of liquid with metal surface. The minimum required KR (96 %) was achieved with the treatment ratios higher than 0.85 g/100 mL within 0.5 h. With 0.73 g/100 mL, 0.65 g/100 mL, and 0.58 g/100 mL ratio, the disinfection was not effective in short retention time. The final KR of the 0.37 g/100 mL and 0.28 g/100 mL groups could reach 96 % and 95 % at 4 h retention time. Comparatively, the 1.10 g/100 mL, 1.00 g/100 mL, 0.92 g/100 mL, and 0.85 g/100 mL treatment ratios had a better killing performance with shorter treatment duration. As the KR values of the four groups were similar during the experiment conducted 0.85 g/100 mL was identified as the optimal treatment ratio for the small-scale CSTR trials due to lower material requirement per unit volume of treated wastewater. However, it was desired to know if this optimum ratio would hold when the reactor volume was significantly increased, as discussed in the next section.

3.3. Process scale-up and batch to continuous piloting

To investigate the scalability of the copper foam treatment process, intermediate-scale laboratory CSTR experiments were first conducted. The optimal metal-to-liquid ratio was carried over from the small-scale optimization, but higher ratios were also tested to investigate whether the kinetics of the KR would change at larger scale. The KR values over time of two different treatment ratios are compared in Fig. 5 for total coliform, fecal coliform, and E. coli. Maintaining the same treatment ratio at 0.85 g/100 mL in intermediate-scale testing, the KR of total coliform and E. coli could only surpass 95 % after 2 h retention time. Consequently, the killing performance of the treatment ratio at 0.85 g/100 mL was not as reliable and effective at the intermediate scale, where the volume was scaled-up by a factor of up to 15.3 (i.e. 10000 mL/650 mL). Therefore, the treatment ratio was enlarged gradually until reaching satisfactory killing performance at 2.00 g/100 mL, namely 97 % at 1 h retention time. This newly optimized treatment ratio was applied in further scaled-up testing, in the continuous pilot-scale experiment, where the process was further optimized.

The pilot-scale experiment was conducted based on the optimized metal-to-liquid ratio obtained from the intermediate lab-scale experiment, at 2.00 g/100 mL. The scale-up factor was 6.4 (i.e. 58 L/91 L) from the intermediate lab-scale experiment to the pilot-scale experiment in terms of reactor liquid volume; however, a new challenge appears: the experiment changes from batch to continuous. Under continuous operation, the copper foam must maintain performance over time. Also, in a plug flow reactor, each metal foam stack contacts wastewater with different bacterial loads: the first stack treats untreated wastewater, while the last stack polishes the treated wastewater. The retention time in such a reactor is a function of reactor length; thus, treated wastewater collected in Valve 1 has had the lowest retention time and at Valve 4 the highest retention time. It is also important to note that such plug reactor requires some time to reach steady-state operation, since the flow pattern during reactor filling differs from the flow pattern developed once the reactor is full, at which time each stack experiences constant wastewater flux. Lastly, the stack design ensures all wastewater contacts the metal foam equally, as wastewater flow enters all available pores, thus maximizing metal/liquid contact. The bacterial counts using the stacked foam design, at each sequen-
Fig. 4. (a) Killing rates of total coliform under different treatment ratios at small lab-scale with WWTP wastewater; (b) Killing rates of fecal coliforms under different treatment ratios at small lab-scale with WWTP wastewater; (c) Killing rates of E. coli under different treatment ratios at small lab-scale with WWTP wastewater.

Fig. 5. Killing rates of total coliforms under different treatment ratios at intermediate lab-scale with WWTP wastewater.
tial reactor valve, over different treatment times, are illustrated in Fig. 6a, b and c.

The pilot-scale reactor was operated for 12 consecutive hours, with 12 samples taken from each valve and from the inflow over the run time. The total volume treated, based on the reactor volume (58 l), run time (12 h) and wastewater flow rate (2.07 l/min) was thus 1,490.4 l. At pilot-scale, instantaneous KR was not calculated, since at any point in time the treated wastewater sampled does not correspond to the wastewater at the inflow, which has variable bacterial load given its continuous supply from the WWTP. Instead, KR should be calculated using time-averaged values of bacterial counts over extended periods. The 12-h mean bacterial counts at the inflow for total coliforms, fecal coliforms and E. coli during the run were, respectively: 492.1, 224.4 and 183.2 CFU/mL. Likewise, these same 12-h mean bacterial counts at valve 4, which is at the outflow, were: 23.3, 5.5 and 4.0 CFU/mL. Taking ratios between mean inflow and mean outflow, the KR value was 95.3 % for total coliforms, 97.5 % for fecal coliforms, and 97.8 % for E. coli. Thus, at pilot-scale, the required minimum KR was met. It should be noted that the residence time, calculated based on flow rate and reactor volume, was only 28.0 min; hence, the pilot-scale reactor performed more effectively than the intermediate lab-scale reactor at the same metal-to-liquid ratio. This is likely due to better metal/liquid contact, for two reasons. One, in terms of all flow passing through the metal foam pores at the pilot-scale, multiple times (given the multiple foam stacks). Two, in terms of the inherent characteristics of the plug flow regime ensuring that all flow experienced similar contact with the metal foam (i.e. tight distribution of reaction experience by every element of flow).

From Fig. 6a, b and c, several process behaviours can be observed. First, the first samples collected at 30 min had considerably higher bacterial load than the next sample collected at 1 h. This means that the reactor had not reached steady-state performance in the first thirty minutes, which is just over one residence time. By the one-hour mark, and over two reactor volumes of flow having passed each sampling valve, it is more likely that the performance of a plug flow reactor has reached steady-state.

Second, the inflow bacterial load varied throughout the experiment, and this oscillatory behavior carried through to all sampling valves, except for being attenuated at valve 4 given that at the outflow the bacterial load was either null or small (< 60 CFU/mL total coliforms, and < 16 CFU/mL E. coli). In fact, the other temporal trend to be seen is the bacterial performance over run time, to detect any decline in performance. This can be quantitatively assessed by fitting straight-line equations to the bacterial count at each valve, and noting the slope of each equation for signs of improving (negative) or declining (positive slope) performance. The slopes for valves 1–4, ignoring the first sample before steady-state is deemed to have been reached, respectively were: 0.559, 2.467, -0.767, and 0.057 CFU/(ml h). Thus, performance slightly decline over time at valves 1 and 2, and improved or remained stable at valves 3 and 4. Notably, the inflow wastewater trended upwards during the 12-h experiment (slope 5.993 CFU/(ml h)), hence it can be concluded that copper foam performance was stable for the run duration tested.

Third, the bactericidal performance consistently improved from valves 1–4; it should be noted again that values at a given point in time should not be compared, since the fluid plugs at each location originate from different fluid plugs entering the reactor, which in turn had variable bacterial load. Hence, the best way to confirm that bactericidal performance is proportional to residence time is to take 12-h mean bacterial counts at each valve and generate 12-h averaged KR values (see Table 1).

In all cases listed in Table 1, the bacterial counts gradually reduce as the wastewater travels down the tubular reactor and passes
through multiple metal foam stacks (two between each sample), resulting in gradually increasing KR along the reactor length. KR values for fecal coliform and E. coli were similar at each valve, while total coliforms KR lagged but caught up by the outflow. This suggests that certain bacterial species that appear in the total coliforms count are more resistant to copper contact, but eventually are impacted. Lastly, it should be noted that the residence time of the pilot-scale tubular reactor was optimized in preliminary trials (the piloting experiments were part of a long campaign of reactor testing and optimization), so residence time, superficial liquid velocity and reactor length, metal-to-liquid ratio, and metal foam positioning and packing within the reactor, are critical design parameters to achieving desired performance.

There is an abundance of studies that have investigated the potency of metal materials on antibacterial applications in a laboratory-scale; however, the feasibility of their scaled-up application remains scarce (Baruah et al., 2016; Maguire-Boyle et al., 2012; Pulit et al., 2013). The present study tested the efficiency of copper foam treating municipal wastewater as well as its stability treating a larger amount of wastewater continuously. Treating municipal wastewater is challenging with many design and performance factors involved, and further technology development is warranted, particularly to test long-term performance and foam cleaning requirements. To be noted from the present study, the plug flow tubular reactor significantly reduced the dead zone volumes, versus typical continuous processing units commonly used in water treatment applications, and improved metal/liquid contact efficiency, versus batch reactors. It is thus recommended that in large-scale applications the tubular reactor design be considered to improve disinfection efficiency. There was no energy supply throughout the pilot-scale experiment, since the water flow was sustained by gravity, which is attractive for large water treatment systems. In fact, a tubular reactor can also be driven by syphoning action when a height difference between inflow and outflow is not possible. This scale-up study showed a promising result of the application of copper foam in wastewater disinfection since it not only demonstrated the prominent antibacterial capability of copper foam but also the low energy consumption and high operating flexibility.

Material characterization was conducted at the end of the experiment to examine the durability of copper foam and to have a better understanding of its interaction with microorganisms (as shown in Fig. 7). It is shown that the interconnected porous structure of copper foam was stable throughout the experiment, which further supports its robustness and stability. However, it is visible that the partial oxidation of copper occurred under the aerobic condition. Though the effects of oxidized copper on the antibacterial performance were not observed and the performance proved stable throughout this study, the potential inhibition of the antibacterial activities in long-term applications still needs to be considered. According to the study of Akhvan and Ghaderi (2010), copper oxide nanoparticles showed a lower inactivation on E. coli compared to intrinsic copper. Therefore, the occasional treatment of surface oxidized copper layer may be needed. Dissolution of copper oxide layer by immersing in mineral acid may be one of the treatment solutions (Habbache et al., 2009); however, it might shorten the longevity of copper foam. The removal of copper oxide will reduce copper mass, and the durability and antimicrobial performance of copper foam may be accordingly impacted. This needs to be determined experimentally in subsequent long-term testing. Another potential treatment strategy is to reduce the oxidized copper to intrinsic copper (Kirsch and Ekerdt, 2001). The reduction may be achieved by using hydrogen, carbon monoxide, etc. This process may need external heating to accelerate the process and that could potentially impact the mechanical strength of the material, and lead to the structural change of the microstructured pores. The antibacterial effects of oxidized copper and detailed long-term maintenance strategy of copper foam will need to be addressed in future work.

In addition to the concern of oxidized copper, biofilms and fouling should also be considered for long-term applications, especially treating wastewater having a high load of residual solids and microorganisms in the tertiary treatment effluent. The maintenance strategies can likely rely on periodic chemical backwash to dislodge foulants and dissolve scale and biofilms. According to the study of Mikolay et al. (2010) on metallic copper surfaces, a solution containing glucoprotamin can be used as a cleaning deterrent, which itself is a disinfectant against microorganisms (Zeitler and Rapp, 2014). The study showed that the cleaned surfaces had lower survival of microorganisms compared to the uncleared surfaces. Harrison et al. (2008) found that the quaternary ammonium compound can work synergetically with ionic copper to eradicate the biofilm of E. coli, Staphylococcus aureus, Salmonella enterica, and Pseudomonas fluorescens. Therefore, chemical detergents may be adopted for cleaning metal foams to eradicate biofilm and facilitate the antibacterial activities, and this should be subject of further study before full-scale implementation.

### 3.4 Leaching risk test

The ICP-MS analysis showed that the prepared bacterial solutions, before metal foam treatment, had copper and nickel concentrations below the detection limit (0.1 µg/L). Fig. 8a compares the leaching levels of copper and nickel in metal foam-treated wastewater samples from small lab-scale experiments. In these batch reactor tests, the retention time was positively correlated to the leaching concentration of copper and nickel up to a solubility limit reached at 3 h. Values at 4 h slightly decreased, possibly due to slight external condition changes such as pH, temperature, and CO₂ fugacity. With the same mass of foam applied, copper had double the maximum leaching concentration compared with
nickel. The maximum concentration of leached copper and nickel was approximately 10.8 μg/L and 4.8 μg/L, respectively. The higher concentration of copper ions correlates with the higher bactericidal efficiency of copper material compared to nickel. However, it should be noted that Sicairos-Ruelas et al. (2019) tested 400 μg/L soluble copper as a wastewater disinfectant, with no significant effect found. Thus, contact with copper foam surface is likely the dominant bactericidal effect over that of copper ions, which is a promising trait for long-term performance of copper foam with minimal material degradation (i.e., leaching) required. Subsequent experiments should be conducted to have a better understanding of the disinfection mechanism of the copper foam.

Fig. 8b illustrates the leaching behavior of copper in the intermediate lab-scale batch reactor. Here, the WWTP secondary treatment wastewater, before metal foam treatment, had an initial copper concentration of 0.16 ± 0.01 μg/L, as copper is common trace component of municipal wastewater as well as natural waters. In the first hour of treatment, copper continuously leached out in the aqueous solution and reached the highest concentration (1.33 μg/L). After the first hour, the concentration settled in the range between 1.0 μg/L to 1.2 μg/L until the end of treatment. Following Canadian water quality guidelines (CWQG), and considering the average hardness of Guelph water (448 mg/L as CaCO₃) (City of Guelph, 2018), the copper concentration should be lower than 4.0 μg/L to protect aquatic life and ecosystem health (Lumb et al., 2006). The leaching risk of copper foam for wastewater disinfection under minimized material use (metal-to-liquid ratio) in the intermediate lab-scale reactor was thus relatively low.

In the pilot-scale experiment, the initial copper concentration in the influent was 0.29 ± 0.01 μg/L. Fig. 8c shows the trend of total copper concentration with increased run time at each valve location. The leaching concentration in treated water was positively correlated to reactor length (i.e., increasing from closest to inflow, valve 1, to closest to outflow, valve 4). Thus, as wastewater passes through each foam stack, it picks up copper. However, the highest outflow value recorded was 2.1 μg/L, and the outflow concentration was relatively stable over the course of the experiment, with some variability attributable to wastewater compositional changes. The continuous flushing in the pilot-scale experiment did not impact the leaching risk, and the copper concentration was maintained within a safe range for treated wastewater outfall.

4. Conclusions

This study was carried out to investigate the bactericidal properties of micro-structured metal materials. The tested metal materials could successfully kill wastewater bacteria at different extents and kinetics. Among nickel foam, copper mesh, and copper foam, the copper foam had the best bactericidal performance. The copper foam was applied in a scale-up study, that went from small to intermediate lab-scale, and subsequently to pilot-scale. Based on optimal conditions at each scale, the scale-up factor from small lab-scale to pilot-scale in terms of reactor volume was 89, while that in terms of foam mass was 211. The intermediate lab-scale experiment led to an adjustment in the optimal metal-to-liquid ratio, and thus the two different scale-up factors used, form 0.85 g/100 mL to 2.00 g/100 mL. The optimization target used was a minimum KR value of 95 % for total coliforms and E. coli. At pilot-scale, a continuous plug flow tubular reactor proved successful in treating multiple times (25.7) the amount of wastewater.
inflow compared to the reactor volume during a 12-h run with 28.0 min residence time. The satisfactory performance reached (96 % KR) was attributed to the reactor design, the operating parameters, the metal foam packing, and the metal foam micro-structure. Thus, the implementation of the "structure" PI strategy was deemed successful up to the scale tested, and further development just focused on ensuring the technical and economical feasibility to disinfec

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