Naturally Occurring Oxidised Ferric Hydroxide as a Coagulant for Pathogen Removal

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Abstract

Pathogens such as protozoan cryptosporidium, are not inactivated by chlorination and can therefore cause public health risks. Ground Water Treatment Plants (GWTPs) that do not have a coagulation step have limited protozoan pathogen removal. The pre-chlorination step in the GWTPs produce ferric chloride, a coagulant, which oxidises to form ferric hydroxide. This project investigates the feasibility of using naturally forming ferric hydroxide as a coagulant to aid in pathogen removal. Biotin coated microspheres were chosen as a non-infectious surrogate to determine log removal as it had comparable characteristic features to Cryptosporidium such as size and zeta potential. Synthetic water was prepared and dosed with ferrous sulphate heptahydrate to obtain 1-6 ppm of iron. Jar test experiments and a filtration column setup are used to verify coagulation and to determine the Log Removal Values (LRVs) of surrogate. This research has shown that there is potential for coagulation with iron and chlorination with a LRV of ~1.5 log. The LRVs achieved will be used for future reference at Water Corporation to design GWTPs accordingly to meet the source risk.

1. Introduction

1.1 Project Background and Current Client Environment

To continuously provide safe drinking water, Water Corporation must comply with several policies and guidelines, including the Australian Drinking Water Guidelines (ADWG). A Microbial Health-based target was added to the Australian Drinking Water Guidelines in 2022 by the National Health and Medical Research Council *(National Water Quality Management Strategy, 2017)*. Health-based targets provide a quantitative measure of the microbial safety of drinking water. With the ADWG's health-based targets, there is a need to demonstrate that each water treatment plant meets the log removal requirements of pathogens to provide safe drinking water to the public.

Pathogens are infectious microorganisms that cause diseases in humans and animals. Certain pathogens like cryptosporidium can be difficult to be inactivated by disinfection with chlorine. When feces from leaky sewer lines, septic systems, livestock operations, or wildlife enter the groundwater, these microbial organisms can enter drinking water bores. For most pathogens,

strong oxidising compounds such as chlorine, chloramine, or ozone (UV) act as disinfectants, inactivating microbial cells via a range of chemical reactions. However, cryptosporidium doesn't get inactivated by disinfection as it has a tough outer layer. This makes it difficult to effectively kill the cryptosporidium oocysts in water. Chemical coagulation is an important factor in determining filtration efficiency. Without pre-treatment, rapid rate filtration works as a simple strainer and is not an effective barrier for pathogens (*Water Treatment and Pathogen Control*, n.d.).

There are seven locations in WA that currently do not have coagulant dosing, solely using chlorination before filtration to oxidise any iron and manganese present in source streams (Surface Water Treatment Manual, 2021). However, as ferric hydroxide is produced through chlorination, the project focuses on the potential of using it as a coagulant. If ferric hydroxide can be proven to be an effective coagulant, then Water Corporation can potentially claim LRVs at these seven locations. The investigation is done on a surrogate, having similar characteristics of cryptosporidium to obtain its Log Removal Values (LRVs).

LRVs give us the concentration of pathogens removed from water as described below:

$$\text{Log removal} = \log (C_{in}) - \log (C_{eff})$$

where C_{in} is the concentration of the species in the feed and C_{eff} is the concentration of the species in the effluent. For example:

- 1 log removal = 90% reduction in density of the organism,
- 2 log removal = 99% reduction,
- 3-log removal = 99.9% reduction etc

1.2 Literature Review

Cryptosporidium spp. detection in water can be difficult due to their low abundance and the complexity of the water matrix (Hassan et al., 2020). As this microorganism imposes a health risk, a surrogate with similar particle characteristics to cryptosporidium is chosen. Some examples of surrogates investigated include bacillus subtilis (Bradford et al., 2016), and aerobic spores (Facile, 2000). Instead of using an infectious substitute, a non-infectious surrogate such as carboxylated microspheres was considered.

1.2.1 Selection of best suitable surrogate

A number of parameters such as size, shape, zeta potential, and density were compared to choose a surrogate between unmodified carboxylated polystyrene microspheres (CPMs), biotin coated, and glycoprotein coated CPMs. Research showed that biotin CPMs are superior in analysing the filtration removal when compared to unmodified CPMs (Stevenson et al., 2015).

1.2.2 Quantification of cryptosporidium surrogate in water

Detection techniques that can quantify the presence of cryptosporidium include microscopy (conventional staining, immunofluorescence assay, etc.), molecular (PCR, qPCR, etc.), cytometry (flow cytometry) and optical (Fluorescent in-situ hybridization (FISH), etc) (Hassan et al., 2020). These detection techniques were compared to choose a reliable analysis for the surrogate. Specificity, sensitivity, repeatability of results, speed, and automation, are the most

significant prerequisites for reliable analysis. These parameters were evaluated for quantifying biotin microspheres, and it was found that flow cytometry is the most reliable and time effective analysis.

1.3 Project Objectives

- To understand whether chlorination oxidises the ferric ions present in water to form ferric hydroxide which can act as a potential coagulant.
- To obtain the Log Removal Values (LRVs) of biotin microspheres using the naturally occurring ferric hydroxide as a coagulant before filtration.
- Use a filtration column mimicking the sand filtration unit to determine LRVs.

The major objective of Water Corporation on this project is to achieve a long-term cost reduction with the existing infrastructure. If proven that ferric hydroxide can be used as a coagulant, the costs expected to be saved at the seven locations would roughly be A$ 1-2 million per year *(Water Corporation, 2022)*.

2. Process

2.1 Experimental Investigations

2.1.1 Preparation of Synthetic Water

5L of synthetic water was made-up using 50 ml of 0.5 mmol/l NaHCO3, 50 ml of 0.3 mmol/l CaCl2, 50 ml of 0.2 mmol/l of MgSO4 and remaining volume of DI water. Synthetic water dosed with ferrous sulphate hypochlorite is used for this study.

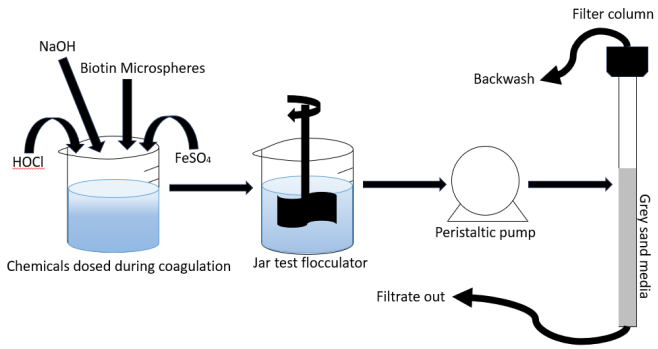

Figure 1 Water treatment setup.

2.1.2 Jar Test

Jar test experiments were performed to verify coagulation and to determine the log removal of surrogate that can be achieved under varying conditions. Throughout the experiments, the dosage concentrations of chemicals were 0.0125% (w/v%) biotin microspheres, 1% (w/v%) ferrous sulphate ($FeSO_4$), 1.04% hypochlorous acid (HOCl) (w/v%), and 0.1M sodium hydroxide (NaOH), which is used to adjust pH. The experiments were performed for three samples: A: Biotin + Synthetic Water; B: Biotin + Synthetic Water + Ferrous Sulphate; and C: Biotin + Synthetic Water + Ferrous Sulphate + Hypochlorous Acid + Sodium Hydroxide. The process steps are:

1. Add 800 ml of synthetic water into a series of jars and dose 0.1 ml of biotin microspheres.

2. Immediately, stir each jar at a constant speed of 120 rpm. After 30 seconds, dose 2.4 mL $FeSO_4$, 0.3 mL HOCl, and 1.5 mL NaOH in order, at 30 seconds interval to three different jars. After dosing, switch the flocculator speed to 30 rpm to ensure uniform distribution of iron.

3. Allow 5 minutes of mixing, observe flocs formation, and rate their floc size in a chart.

4. As per industry jar test standards, Whatman grade 1 filter papers of 11 µm and a standard filter column were used. Filtered samples were analysed using flow cytometry.

2.1.3 Design of filtration column

The filter column was designed to replicate existing filters to understand whether the filtration technique will work in full scale plants. A glass column with dimensions 50 × 3 × 0.2 cm was chosen to hold the filter media. Grey sand with a size range of -1.0 + 0.5 mm was chosen as the filter media and is filled in the column to a bed height of 20 cm. Inlet and outlet tubes are connected by valves for samples to enter and exit the column, with a provision for backwash. A peristaltic pump is used to control fluid flow through the column. For filtration the flowrate set is 100 mL/min and for backwash, it is 340 mL/min. The sand must be cleaned regularly by backwashing for 10 minutes to maintain the filtration efficiency. The filtered samples were analysed using flow cytometry and the results were compared with the filter paper tests.

2.1.4 Data analysis

Flow cytometry analysis is used to detect the microspheres and determine the concentration. The fluorescence emitted by the biotin microspheres is collected by detectors and converted into electrical signals, which are processed by a computer. All samples were analysed for 4 mins to allow more detection of biotin microspheres. The instrument gives the count of microspheres detected through fluorescence and classifies the results as single layered and double layered particles, where the double layered proved coagulation occurred. By obtaining the concentrations of samples from pre-filtration and post-filtration, the LRVs of the microspheres can be calculated.

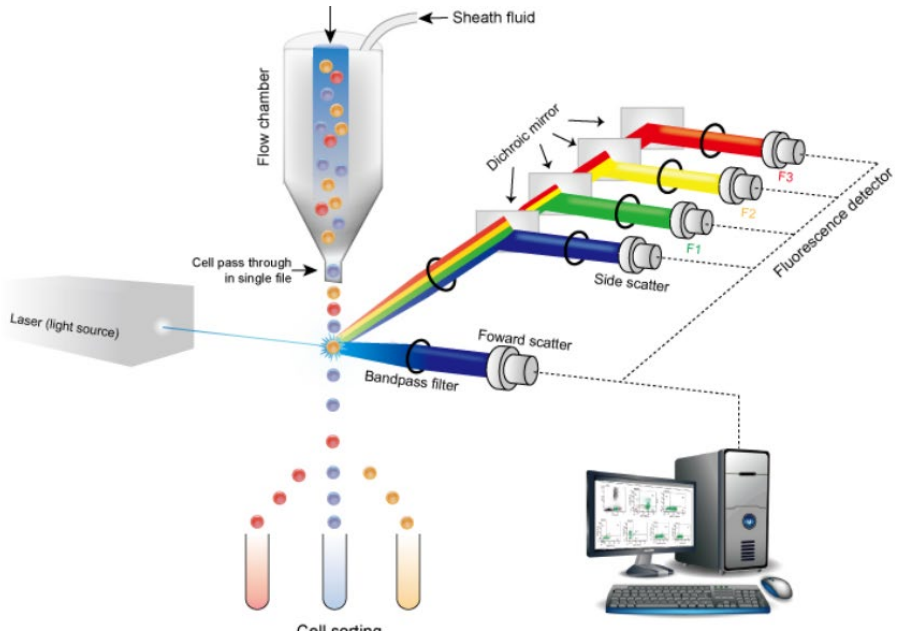

Figure 2 Cell Detection in flow cytometry (Rowley, 2020)

3. Results and Discussion

The results from the jar tests with filter papers showed that the biotin microspheres in synthetic water were filtered without even dosing ferrous and/or chlorine. To find the reason for filtration of microspheres in a large 11 um filter paper, Scanning Electron Microscopy (SEM) analysis was performed to check the particle size and the average size found was 5.3 µm. It was then decided to repeat the same experiments in a filter column to check whether the biotin microspheres pass through the filter media. A few trials performed in filter column with samples A and C (mentioned in section 2.1.2) were analysed in flow cytometry.

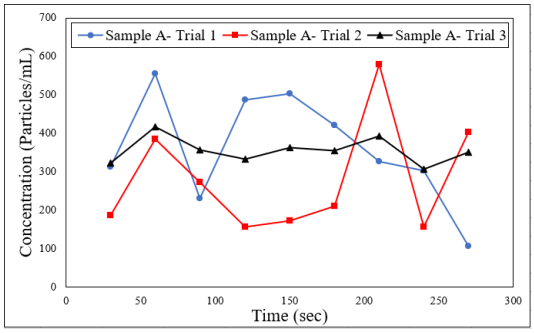

Figure 3 Uncoagulated biotin microspheres leaving filter column at every 30 seconds

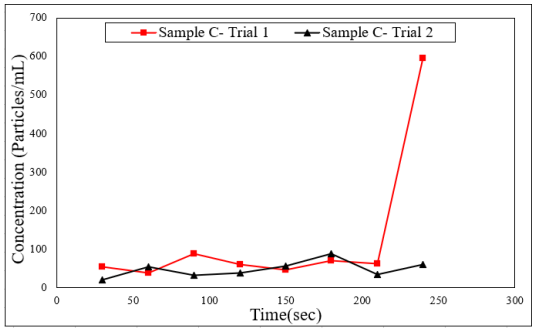

Figure 4 Coagulated biotin microspheres leaving filter column at every 30 seconds

As observed in Figure 3, biotin microspheres in synthetic water passed through the filter column unlike the filter paper tests. Figure 4 depicts the coagulated microspheres that had passed through the filter column. It can be observed that the concentration of microspheres in Figure 3 is greater than the ones in Figure 4, which proves that the microspheres have coagulated with the dosage of chemicals. To further verify whether ferric hydroxide had played a major role in coagulation, zeta potential analysis would be performed on samples A, B, and C. At every instance, the filtrated sample has varying concentrations of biotin microspheres as shown in Figure 3 and 4. To obtain the log removal of biotin microspheres, a sample collection time has to be set after which the particles ejected are consistent. A trial experiment was performed to verify this.

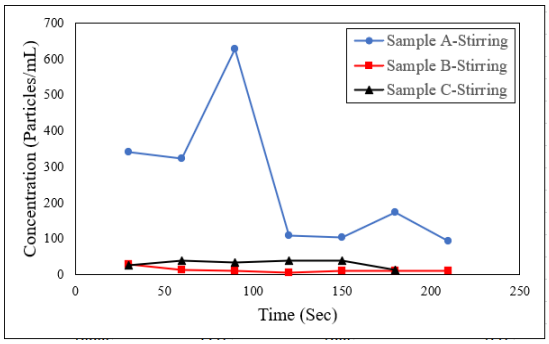

Figure 5 Trial experiments with samples A, B, and C, continuously mixed during filtration.

Figure 5 depicts that the particle removal is more consistent with mixing, which will allow us to collect the sample at any instance after filtration. However, as the particles coming out of filter column in sample A is still fluctuating, a few more trials will be performed to identify the sample collection point. Comparing figures 3, 4, and 5, it can be shown that the particles are coagulated, and the flocs are dispersed unevenly in the samples.

4. Conclusions and Future Work

Filter paper tests haven't shown promising results, but the few experiments performed using filter column are showing good removal with ferric hydroxide. The results in filter column experiments have shown that the particles have coagulated with ferric and chlorination. However, to confirm whether ferric hydroxide has played a major role in coagulation, zeta potential of the samples would be analysed to understand the charge neutralisation. Preliminary results show ~1.5 log removal is achieved for the experiment with continuous mixing for samples B and C. Further, log removal values will be obtained once the sample collection point is set.

5. Acknowledgements

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6. References

Bradford, S. A., Kim, H., Headd, B., & Torkzaban, S. (2016). Evaluating the Transport of Bacillus subtilis Spores as a Potential Surrogate for Cryptosporidium parvum Oocysts. Environmental Science & Technology, 50(3), 1295–1303. https://doi.org/10.1021/acs.est.5b05296

Facile, N. (2000). Evaluating bacterial aerobic spores as a surrogate for Giardia and Cryptosporidium inactivation by ozone. Water Research, 34(12), 3238–3246. https://doi.org/10.1016/s00431354(00)00086-5

Hassan, E. M., Örmeci, B., DeRosa, M. C., Dixon, B. R., Sattar, S. A., & Iqbal, A. (2020). A review of Cryptosporidium spp. and their detection in water. Water Science and Technology, 83(1), 1–25. https://doi.org/10.2166/wst.2020.515

National Water Quality Management Strategy. (2017). https://www.nhmrc.gov.au/sites/default/files/documents/reports/aust-drinking-water-guidelines.pdf

Rowley, T. (2020). Flow Cytometry - A Survey and the Basics. Materials and Methods. [online] Available at: https://www.labome.com/method/Flow-Cytometry-A-Survey-and-the-Basics.html.

Stevenson, M. E., Blaschke, A. P., Toze, S., Sidhu, J. P. S., Ahmed, W., van Driezum, I. H., Sommer, R., Kirschner, A. K. T., Cervero-Aragó, S., Farnleitner, A. H., & Pang, L. (2015). Biotin- and Glycoprotein-Coated Microspheres as Surrogates for Studying Filtration Removal of Cryptosporidium parvum in a Granular Limestone Aquifer Medium. Applied and Environmental Microbiology, 81(13), 4277–4283. https://doi.org/10.1128/aem.00885-15

Water Treatment and Pathogen Control. (n.d.). http://apps.who.int/iris/bitstream/handle/10665/42796/9241562552.pdf

Surface Water Treatment Manual (2021). Water Corporation, 1-48.