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Identification of the Chlorine- and Bromine-Based Biocides - Task 1 of the NESC Assessment of Biocide Impacts on Life Support (LS) and Extravehicular Activity (EVA) Architectures

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National Aeronautics and Space Administration (NASA) has undertaken an Assessment of Biocide Impacts on Life Support (LS) and Extravehicular Activity (EVA) Architectures, producing an NASA Engineering and Safety Center report by the same name. The purpose of the report is to evaluate whether the current iodine-based biocide should be replaced due to the need of removing that iodine prior to consumption, if silver biocide is an appropriate replacement, or if there is another better biocide choice. This paper is a supporting document detailing the chlorine- and bromine-based biocides found for Task 1 (Identify and Document Available Forms of Biocidal silver, bromine, and chlorine). This paper identified eleven chlorine-, eight bromine-, and one mixed chlorine- and bromine-based biocides and documented their many chemical, biological, and application properties. Additionally, five qualification criteria were developed to evaluate the biocide's acceptability. Based on this evaluation, ten of the twenty biocides found were not recommended for further evaluation in Tasks 2-4, six are being recommended after passing all qualification criteria and four are being recommended requiring further review.

Nomenclature

Ag^+ = silver ion
BCDMH = 1-bromo-3-chloro-5,5-dimethylhydantoin
Br = elemental bromine
Br_2 = bromine
BrO^- = hypobromite ion
Cl = elemental chlorine
Cl_2 = chlorine
ClO^- = hypochlorite ion
ClO_2 = chlorine dioxide
DBDMH = 1,3-dibromo-5,5-dimethylhydantoin
DBNPA = 2,2-dibromo-3-nitrilopropionamide
EPA = Environmental Protection Agency
EVA = extravehicular activity
FDA = Food and Drug Administration
HBr = hydrobromic acid
HCl = hydrochloric acid

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HOBr = hypobromous acid
HOCl = hypochlorous acid
ISS = International Space Station
I_2 = iodine
LS = life support
NaDCC = sodium dichloroisocyanurate
NASA = National Aeronautics and Space Administration
NH_2Cl = monochloramine
ppm = parts per million
WHO = World Health Organization
xEMU = Exploration Extravehicular Mobility Unit

I. Introduction

As National Aeronautics and Space Administration (NASA) turns to Exploration missions beyond low-Earth orbit, these missions reflect new challenges in regards to water treatment and maintenance for long usage and dormancy periods. Iodine (I_2) is currently the biocide used within the United States Orbital Segment of the International Space Station (ISS) and has a long history of use in space missions dating back to the Apollo missions within wetted portions of life support (LS) and extravehicular activity (EVA) systems.[1] However, because of challenges with compatibility and dormancy, the LS Systems Water Processing Team has elected ionic silver (Ag^+) as a biocide for all Exploration missions. Ag^+ can be safely consumed at biocidally active concentrations as opposed to I_2, but it poses challenges in terms of dosing and material compatibility as ionic silver readily reacts with the materials in the water system.[1] The EVA Systems Water Processing Team has selected I_2 as a biocide for ISS demonstration and Exploration missions. Because of the inherent interfaces between the Exploration Extravehicular Mobility Unit (xEMU) and LS water, significant concerns exist regarding the interactive effects of these chemicals and their effects on materials and hardware. As a result, there is a demand for a single biocide solution that can remain effective in multiple materials and that does not impact the water system hardware. With these challenges for the biocide, there is an opportunity for bromine (Br) and chlorine (Cl) biocides to be considered in future Exploration missions.

The purpose of this literature study was to identify and document available forms of biocidal Br and Cl and to assess their potential use in spacecraft potable water systems as a part of Task 1 of a NASA Engineering and Safety Center Technical Assessment Report of all available biocide solutions.[2] This study identified eleven chlorine-, eight bromine-, and one mixed chlorine- and bromine-based biocides. These biocides were assessed based on the five Qualification Criteria listed in Table 1.[2] If the biocide fails any of the criteria (Option 1), it was disqualified from further assessment. If not enough information or studies was obtained to make an assessment on a criterion, it was noted as an "Unknown" and flagged for additional evaluation to the other Task groups (Option 2). If the biocide fulfills all five criteria (Option 3), it passes our assessment and qualified for evaluation in Tasks 2-4 (crew health effects, xEMU compatibility, and LS hardware compatibility, respectively). Our assessment found six Cl and Br biocides that were recommended for further evaluation to Tasks 2-4 based on their fulfillment of these criteria. A table summarizing the assessments of the 20 biocides based on the Qualification Criteria can be found in the Appendix.

Table 1. Biocide Qualification Matrix. Examples of outcomes are provided for reference only.[2]

Criteria	**Opt 1**	**Opt 2**	**Opt 3**
Must not introduce a known risk to crew health at effective biocide concentrations	🟩	🟩	🟩
Must not require more than 3 years to begin flight demonstration/implementation	🟩	🟩	🟩
Must be commercially available or can be made available quickly for evaluation in NASA ground systems.	🟩	🟩	🟩
Must not be a Toxic Hazard Level 3 (storage or use)	🟥	🟨	🟩
Must not degrade/become ineffective under storage conditions (non-use) for up to 3 years	🟩	🟩	🟩

Determination	Disqualified	Obtain additional data	Qualifies for Tasks 2-4

🟩	**PASS**
🟨	**UNKNOWN**
🟥	**FAIL**

II. Bromine and Chlorine-Based Disinfection

Most chlorine- and bromine-based biocides are biocidal due to their oxidative nature. The biocidal species which they produce are the hypochlorous (ClO^-) and hypobromous (BrO^-) ions and their associated acidic forms (HOCl and HOBr, respectively) (Eq. 1). These biocides are formed from one of two methods. The first is the solvation of an ionic salt, sodium hypochlorite (NaOCl) for example, to release the hypohalite ion (Eq. 2). The second is the hydrolysis of a halogen-containing biocidal species to form the hypohalite in the acidic form (Eq. 3).

$$HOX \leftrightarrow H^+ + XO^- \; ; \; X = Cl \; or \; Br \tag{1}$$

$$MOX \leftrightarrow M^+ + XO^- \; ; \; M = monovalent \; cation, X = Cl \; or \; Br \tag{2}$$

$$RX + H_2O \rightarrow RH + HOX \; ; \; X = R = Cl, Br, or \; organic, X = Cl \; or \; Br \tag{3}$$

It should be noted at this point that the ionic forms can be written as either XO^- or OX^- with the equivalent being applied to the molecular forms as well. The difference is whether the IUPAC or industry standard is being applied.

These hypohalite species are capable of oxidizing biomolecules within bacteria and viruses. With a 4 ppm dose of HOBr, experiments have observed a 3.7-5.0 log reduction of *E. coli* and 3.7-4 log reduction of f2 phage virus in pH 7 water.[3] For HOCl, 0.02 ppm of the biocidal species was enough to achieve a 2 log reduction of *E. coli* in pH 7 water.[4] As a result, Cl is approved for potable water disinfection applications with a 5 ppm guideline level set by the World Health Organization (WHO)[5] and a 4 ppm maximum concentration allowed by the Environmental Protection Agency (EPA)[6]. Br, while approved for potable water disinfection, has not seen as much use in potable water systems in comparison to chlorine. This is partly due to concerns over bromine disinfection by-products.[3] However, Br has been implemented in the U.S. Navy in their vessels' potable water systems with their guidelines requiring 0.2 ppm minimum residual Br concentration for disinfection.[7] The EPA has set the maximum concentration of the residual Br to 1.0 ppm in the final treated water.[8]

While halogen-containing biocides are effective due to their oxidative power, that same efficacy is a primary issue of concern. Their oxidative characteristic makes them extremely corrosive to many materials, including metals. The acidic form, HOX, is generally considered to be more corrosive than the ionic form XO^- due to the pH dependence of the electrochemistry.[9] This is an issue because the potable water on the ISS is around pH 5.[10] The potable water of future exploration missions is expected to be the same. HOCl has a pK_a of 7.4 and HOBr has a pKa of 8.5. At pH 5, HOBr and HOCl are the predominant species in water as the pH is well below their pK_as. As a result, the biocide remains at the more oxidative form that can affect both bacteria and metals within the potable water system.

Another potential issue with Br and Cl disinfection is the formation of potentially hazardous by-products. These by-products include bromate (BrO_3^-) and trihalomethanes,[11] which are carcinogens with EPA maximum concentration limits of 10 ppb[5] and 80 ppb[5], respectively. The issue of hazardous by-product formation was brought up to the Task 3 Crew Health Effects group for further review.

Finally, XO^- is degraded by heat and light.[12] These issues, although minimized by the current water processing and xEMU designs, can still be problematic due to the long storage life requirement of Exploration missions.

III. Biocide Properties and Qualification Assessment

A. Gases

The biocidal halogen gases and related gas compounds are bromine (Br_2), chlorine (Cl_2), bromine monochloride (BrCl), and chlorine dioxide (ClO_2). Gases can be dosed easily by bubbling into the water supply and controlling the flow rate to adjust the concentration.

Br_2 is a high vapor pressure, reddish-brown liquid at room temperature (212 mmHg vapor pressure at 20 °C) that readily vaporizes and reacts in water to form HOBr and hydrobromic acid (HBr). Based on trends in the periodic table in relation to electronegativity, bromine is known to be more reactive than iodine and less reactive than chlorine.[11] Br_2 is approved for use in disinfection by the EPA to purify drinking water aboard U.S. Naval ships and offshore oil well platforms at concentrations of 1 ppm within the final treated water.[8] A study by Koski showed that 0.56 ppm Br_2 added to unbuffered water can reduce 6 log *E. coli* to less than 1 log within 30 seconds.[13] However, Br_2 is known to be very corrosive and can cause pitting and damage to most metals other than titanium and 28% chromium/3% molybdenum ferritic stainless steel.[14]

Cl_2, a pale-green gas, reacts in water to form HOCl and hydrochloric acid (HCl). Cl_2 is approved as a potable water disinfectant with an EPA maximum limit of 4 ppm.[6] The biocidal study by Koski showed that 0.30 ppm Cl_2 added to unbuffered water can reduce 6 log *E. coli* to less than 1 log within 30 seconds.[13] Cl_2 has been used in municipal potable water applications in the past, but the use of NaOCl is preferred due to the health risks.[15]

BrCl, a highly reactive yellow gas, reacts with water to form HOBr and HCl (Eq. 3). The gas is primarily used for industrial water treatment with the EPA recommending 10 ppm for use in wastewater treatment.[16] A study by Hajenian using BrCl on sewage effluent found that 1 ppm BrCl was capable of a 4 log kill of f2 coliphage at pH 6 while Cl_2 required 10 ppm for the same result.[17] The gas is highly unstable and can decompose into Br_2 and Cl_2.[18]

Lastly, ClO_2, a yellow-green gas, has been used in hospital and municipal water treatment applications[19] with the EPA approving its use as a disinfectant in concentrations up to 800 ppb[6]. ClO_2 shows promise over Cl_2 as its biocidal activity is less affected by alkaline conditions than Cl_2. This was demonstrated in a study by Benarde that shows 0.25 ppm ClO_2 can cause over 2 log kill of *E. coli* within 1 minute at pH 6.5 and within 15 seconds at pH 8.5.[20] In contrast, Cl_2 at the same concentration required 30 seconds at pH 6.5 and 300 seconds at pH 8.5 to achieve over 2 log kill of *E. coli*.

However, gases pose a significant toxicity and asphyxiation risk as they are volume-filling within the crew cabin area if they were to leak during storage. As a result, these chemicals fail to meet Qualification Criterion 4.

B. Salts

All the disinfecting salts that were found in this study were chlorite or hypochlorite salts. While hypobromite salts exist in theory, they are rarely isolated and are known to decompose rapidly.[21] The 3 disinfecting salts that were focused on in this study were sodium hypochlorite (NaOCl), calcium hypochlorite ($Ca(OCl)_2$), and sodium chlorite ($NaClO_2$).

NaOCl is readily found in bleach and other commercial detergents and has been approved for emergency potable water disinfection by the EPA (up to 6-8 ppm)[22]. However, the compound is highly unstable and can decompose explosively when stored in the anhydrous state. Sodium hypochlorite pentahydrate can be used, which is more stable and does not pose an explosive hazard. However, the pentahdydrate form is known to rapidly decompose within 1 month when stored at 20 °C and requires storage at less than 7 °C.[23] As a result, NaOCl shows little promise for spaceflight due to the explosion risk of its anhydrous state and the instability of its pentahydrate state.

$Ca(OCl)_2$ is a more stable hypochlorite salt with twice the active Cl content with 2 moles of HOCl released for every mole of compound. It is readily available from multiple commercial sources and has a shelf life of 3-5 years. $Ca(OCl)_2$ is approved for emergency use in potable water disinfection by the EPA (up to 5 ppm).[22] $Ca(OCl)_2$ can induce a 6 log kill of *E. coli* at 0.2 ppm concentration with a 10 minute contact time.[24]

$NaClO_2$ differs from the other hypochlorite salts. It reacts in water to form ClO_2. ClO_2 can then further react with water to produce ClO^- as presented in Section III. A. The WHO has set guideline limits for $NaClO_2$ of 0.7 ppm[5] while the EPA has approved usage up to 1 ppm.[6] Unlike NaOCl, $NaClO_2$ is highly stable and shows no loss after 10 years in storage.[25]

As these salts cannot be polymerized, the only available methods of implementation in spacecraft water systems are to: 1) allow the salt granules to dissolve over time in the water system (either as free granules or embedded granules in a solid matrix), or 2) to use a concentrated salt solution that can be dosed into the water system. However, the first solution poses problems in terms of the fine particles that can escape and then flow freely through the water system. The second solution poses problems with the mass/volume requirements in transporting and storing the concentrated solution. As a result, salts face a major challenge in spacecraft applications and in fulfilling Qualification Criterion 2.

C. Hydantoins

Hydantoins are five-membered, nitrogen-containing ring compounds as shown in Figure 1. Biocidal Cl and Br can be bound to the nitrogen groups of these compounds as found in 1-bromo-3-chloro-5,5-dimethylhydantoin (BCDMH, Figure 1) and 1,3-dibromo-5,5-dimethylhydantoin (DBDMH, not shown).

BCDMH is a solid biocide that releases HOBr and HOCl in water (Eq. 3). BCDMH meets NSF/ANSI 60 standards, and has been approved for use in potable water with a maximum allowable concentration of 9 ppm.[26] BCDMH is effective at controlling bacteria populations with a 1 ppm dose resulting in a 2 log reduction in total bacteria population in a cooling water system observed over 4 days.[27] Another study of BCDMH in wastewater applications showed a 2.8 log reduction in *E. coli* in wastewater with a 6 ppm dose of BCDMH after 5 minutes.[28] The compound is commercially available at both Enviro Tech and Sigura and is known to have a shelf life of 3 years. However, no known potable

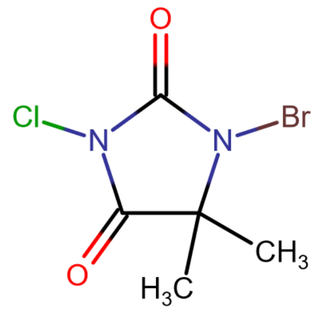

Figure 1. Chemical structure of 1-bromo-3-chloro-5,5-dimethylhydantoin (BCDMH)

water treatment systems currently use this compound. Therefore, its implementation into flight trials within 3 years is unknown. Furthermore, BCDMH in pool applications can cause contact dermatitis[29] and a health risk assessment is required as it is unknown if it has a Toxic Hazard Level (THL) lower than 3 for Qualification Criterion 4.

DBDMH is another solid hydantoin biocide that has been primarily used in recreational and industrial water disinfection. DBDMH, at 1 ppm, was found to be effective at controlling *Legionella* bacteria populations for 24 hours.[30] DBDMH has over a 3 year shelf life and is readily available from Millipore Sigma. However, no potable water treatment system is known to use this biocide, which makes its implementation into flight trials within 3 years unknown.

One significant benefit of these hydantoin compounds is their immobilization by incorporation into a polymer chain. This reduces the challenges of dosing and monitoring the biocidal species concentration levels and provides on-contact kill capability. Hydantoins can be immobilized by means of binding the C5-carbon to a readily-polymerizable monomer like styrene. One commercial product with successful implementation of hydantoins into a polymer is HaloPure BR by HaloSource. The product, a poly-1-bromo-5-methyl-5(4'-vinylphenyl)hydantoin resin cartridge, was based on a similar resin developed by Dr. Worley of Auburn University that can cause >6.8 log reduction of *S. aureus* and *E. coli* after 1 second of contact time.[31] The bromine contained within the resin (16% by weight) kills bacteria on contact and releases residual HOBr/BrO$^-$ at concentrations of 0.1-1.0 ppm, depending on the cartridge design, testing conditions, and shelf life of the HaloPure BR resin.[32] Experiments on HaloPure BR bacterial control in flow-through applications showed a 5 log reduction in bacteriophage MS2[33] and over 5 log reduction of *E. coli* in sewage-contaminated water[34] in the effluent relative to the influent. It is not known how long it will take for HaloPure BR to be implemented in flight testing, but as it is already a resin product with customizable-dimension cartridges, it is expected to be easily plugged in after some initial testing. HaloPure BR is stated to have a shelf life of 2 years when stored with desiccant, which is below the 3 year shelf life requirement of Qualification Criterion 5. This requires further testing to prove if the product can meet this requirement.

D. Isocyanurates

Isocyanurates are six-membered, nitrogen-containing ring compounds that exist in salt and acidic forms as shown in Figure 2. Oxidative Cl is bound to the nitrogens, resulting in chloroisocyanuric acid, dichloroisocyanuric acid, and trichloroisocyanuric acid with higher chlorine content isocyanurates existing in equilibrium with their lower Cl content versions.[35] In theory, Br is another potential biocidal species that could be bound to isocyanurate, but presently no commercially available bromoisocyanuric biocide has been found. Chloroisocyanurates release ClO$^-$/HOCl along with cyanuric acid in aqueous solution. The cyanuric acid serves to stabilize the active Cl species, particularly against UV degradation.[36] However, it also decreases the pH and affects measuring and monitoring of the ClO$^-$/HOCl concentration.

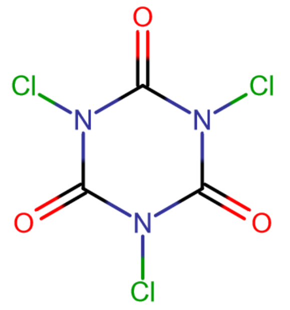

Figure 2. Chemical structure of trichloroisocyanuric acid.

Chloroisocyanurates have extensive usage in drinking, recreational, and wastewater treatment. Two frequently-used chloroisocyanurates are trichloroisocyanuric acid and sodium dichloroisocyanurate (NaDCC). Dissolved trichloroisocyanuric acid exists in equilibrium with di- and mono-chloroisocyanuric acid. As a result of this equilibrium chemistry, trichloroisocyanuric acid gradually releases active Cl over time.[37] The compounds have been approved by NSF/ANSI 60 standards for potable water with a maximum allowable concentration of 30 ppm.[38] NaDCC is the sodium salt version of dichloroisocyanuric acid. It has been commercially available as Aquatabs by Medentech. A biocide study by Schlosser found a 3.5 ppm NaDCC addition led to a 1.8-2.8 log reduction of total bacteria count after 30 minutes of contact time.[39] Both chloroisocyanurates will require further development to be flight-ready, which calls into question their ability to fulfill Qualification Criterion 2.

One significant issue with the use of chloroisocyanurates in potable water treatment is the strong odor and taste of the released Cl. The taste thresholds are highly dependent on the design of the study, but one study determined the taste threshold to be 0.73 ppm.[40] The taste effect of chloroisocyanurates and other halogen-containing biocides is a potential point of concern for crew health that has been referred to Task 2.

Like hydantoins, isocyanurates present another possible polymerizable solution through covalent bonding to the polymer with the nitrogen, but no commercial options were found.

E. Quaternary ammonium salts

Quaternary ammonium salts containing Br and Cl can also act as water treatment disinfectants. However, distinctions need to be made in regards to the active biocide species as quaternary ammonium also acts as a disinfectant. Domiphen bromide shown in Figure 3 is an example of one such quaternary ammonium Br compound that was found to be outside of the scope of this work as the active biocide is the quaternary ammonium, not the bromide ion. However, quaternary ammonium trichloride and tribromide do release active Br/Cl biocide species as the trihalide ion dissociates into the halogen gas and halide ion shown in Eq. 4. The halide gas can then react with water and hydrolyze to form the hypohalite active biocide species as demonstrated in Eq. 3.

$$X_3^- \rightarrow X^- + X_2\ ;\ X = Cl\ or\ Br \tag{4}$$

Umpqua Research Company, which has demonstrated success in the development of the iodine microbial check valve (MCV) on the ISS, is approved for a Phase II SBIR project referred to as Halogen Binding Resin.[41] This resin is similar to the MCV resin with a quaternary ammonium tribromide acting as the biocidal precursor. This work is proprietary and cannot be discussed in further detail, but current findings have found similar active biocide releasing performance to the iodine MCV. As this technology is being developed for space applications, it is considered able to fulfill the flight-readiness requirement of Qualification Criterion 2. However, storage studies would need to be completed to prove that it can remain effective for 3 years as per Qualification Criterion 5.

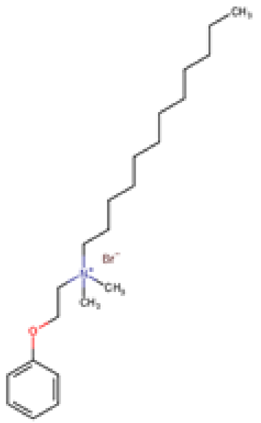

Figure 3. Chemical structure of domiphen bromide.

F. Other Chlorinated Amines

Various chlorinated amine compounds have been used in potable water disinfection applications. Monochloramine (NH_2Cl), a colorless unstable liquid, is an example of the simplest compound of this class. The active biocide of NH_2Cl differs from other oxidizing Cl biocides in that NH_2Cl itself acts as a biocide although it is a less effective viral disinfectant than HOCl.[42] The EPA approves its usage in potable water disinfection at concentrations up to 4 ppm.[6] It has a long history of use in potable water disinfection with it being used in hospital and municipal water systems.[43-45] However, it has a short shelf life during storage, thus requiring *in situ* generation through a reaction between ammonia and chlorine gas, two reactive and toxic gases that would pose a risk to spacecraft equipment and crew.[46] As a result, NH_2Cl fails Qualification Criterion 5.

Halazone, or 4-((dichloroamino)sulfonyl)benzoic acid, is another chlorinated amine solid compound shown in Figure 4A. It was used for point-of-use disinfection of water during World War II with recommended dosages of 4 ppm.[47] However, Halazone has a short shelf life of 5-6 months when left unopened and 3 days when opened,[47] which fails Qualification Criterion 5.

Chloramine-T (Figure 4B) is another chlorinated amine with a structure similar to that of Halazone. It has been approved for the emergency sanitation of potable water as the product Axcentive. Fungicidal testing by de Castro found that Chloramine-T was a more effective fungicide than NaOCl.[48] Chloramine-T also has a shelf life of 3 years.[49]

N-Chlorosuccinimide, has a structure similar to hydantoins as shown in Figure 4C. Early research indicates its potential for potable water treatment with a study by Wood that found 5 ppm of n-chlorosuccinimide was capable of a 3.7 log reduction in bacterial growth after 24 hours.[50] However, no other studies were found that proved this application further, which may impact its fulfillment of Qualification Criterion 2. Polymerization should be possible much like how hydantoins can be integrated in resins.

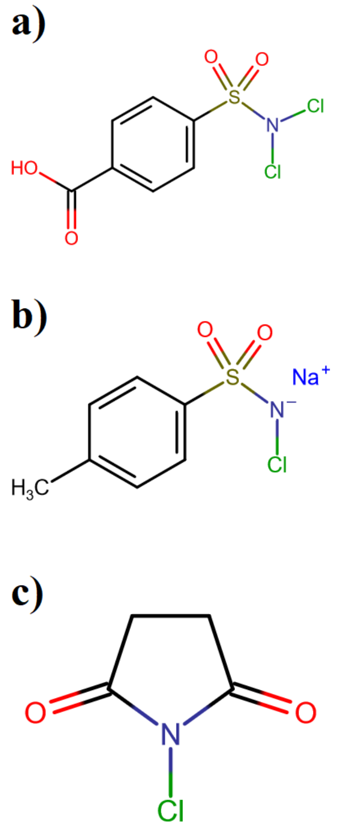

Figure 4. Chemical structures of a) Halazone, b) Chloramine-T, and c) Succinchlorimide.

Both n-chlorosuccinimide and Chloramine-T were used as point-of-use solutions for disinfection and remain unproven in the treatment of potable water systems. As a result, they both require further development to be ready for flight demonstration, leaving their fulfillment of Qualification Criterion 2 unknown.

G. Non-Oxidizing Biocides

In addition to the oxidative Br and Cl biocides investigated, two non-oxidizing Br biocides (Figure 5) were found, 2,2-dibromo-3-nitrilopropionamide (DBNPA) and 2-bromo-2-nitropropane-1,3-diol (Bronopol). The benefit of non-oxidizing Br biocides is they do not cause oxidative damage to materials, preventing issues with corrosion of metallic water system. Both biocides dissolve in water and kill bacteria without requiring additional reactions or solvation to release biocidal species. Studies showed that 0.5 ppm Bronopol caused around 3 log kill of *E. coli* in pH 7 phosphate-buffered water.[51] Addition of 1 ppm DBNPA results in a 2 log kill of *E. coli*[52] and a 2 log kill of *P. aerignosa*[53] in water. DBNPA has stability issues as it has a short half-life of 9 hours when in solution.[53] However, these biocides are only approved in industrial water treatment and not for potable water treatment. As a result, these biocides require Food and Drug Adminstration (FDA) approval, which significantly impacts their readiness for flight, thus failing Qualification Criterion 2. As the Toxic Hazard Level is dependent on FDA testing, these biocides also fail Qualification Criterion 4.

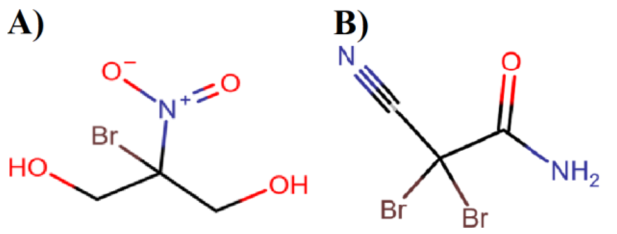

Figure 5. Chemical structures of: A) 2-bromo-2-nitropropane-1,3-diol and B) 2,2-dibromo-3-nitrilopropionamide.

IV. Conclusion

Task 1 of the Assessment of Biocide Impacts on LS and EVA Architectures study was the identification and documentation of available forms of Ag-, Cl-, and Br-based biocides. Biocides from Task 1 were recommended for further evaluation in Tasks 2-4 to identify and document the biocides' effects on crew health, xEMU hardware, and LS hardware, respectively. Task 1 identified eleven Cl-, eight Br-, and one mixed Cl- and Br-based biocides. Their chemical, biological and application properties have been noted and evaluated against the Qualification Criteria of Table 1. From this evaluation, two major findings are noted. First, oxidative Cl and Br biocides have similar biocidal mechanisms of producing either the hypochlorite or hypobromite ion (and their associated acidic form), which are the active biocidal species. Second, of the 20 candidate biocides, six met all of the Qualification Criteria and were recommended for further evaluation in Tasks 2-4, four met some of the Qualification Criteria, but had issues requiring further investigation, and ten failed at least one of the Qualification Criteria and were not recommended.

The isocyanurates (NaDCC, di-/trichloroisocyanuric acid) and the hydantoins (BCDMH, DBDMH, and HaloPure BR), and Umpqua's Halogen Binding Resin met all Qualification Criteria and were recommended for evaluation within Tasks 2-4. Most of the recommended biocides were considered "Unknown" for Criterion 2 due to the uncertainty regarding the amount of time to develop a biocide dosing system with the current flight water system. The only exception to this is the Umpqua's Halogen Binding Resin as that falls under an SBIR project that has already reached Technology Readiness Level 3 with their completion of their Phase I SBIR. Another criterion that has been assessed as "Unknown" for most of the recommended biocides is Criterion 4 as we are unable to make an assessment of the Toxic Hazard Levels of the biocides that would need to be dosed into the water system. The immobilized biocides (HaloPure BR and Halogen Binding Resin) were considered a "Pass" for Criterion 4 as they would only be releasing active $HOBr/BrO^-$ species within the water system. BCDMH, DBDMH, NaDCC, and di-/trichloroisocyanuric acid can be immobilized by polymerization.

$Ca(OCl)_2$, $NaClO_2$, n-chlorosuccinimide, and Chloramine-T all meet several of the Qualification Criteria, but had at least one issue each that required further investigation. $Ca(OCl)_2$ and $NaClO_2$ lack a means of immobilization to a resin and therefore have challenges with an appropriate dosing system for flight. N-Chlorosuccinimide and Chloramine-T require further testing for potable water treatment to prove their efficacy for this application.

All the gases (Cl_2, Br_2, ClBr, and ClO_2) were disqualified due to Criterion 4 as all the gases were toxic and any leak of a gas is an asphyxiation risk to the crew.

Halazone, NH_2Cl, and NaOCl were known to have short shelf lives and, as a result, they failed to fulfill Criterion 5.

The non-oxidizing biocides, DBNPA and Bronopol, were untested and unapproved for human consumption and require FDA testing. As a result, they failed to fulfill Criteria 2 and 4 due to the FDA testing being a major barrier for flight implementation and for Toxic Hazard Level assessment.

The six biocides passing the assessments of Task 1 will continue to be evaluated in terms of effects on crew health and on LS and EVA system hardware. Through these evaluations, NASA can assess the strengths and weaknesses of the biocides on the market today and can make a more informed decision on the biocide solution that fulfills the needs and challenges of Exploration missions in the future.

Appendix

***Table 1. Results of Qualification Criteria on Bromine- and Chlorine-based Biocides*[2]**

Option	**Criteria**				
	Must not introduce a known risk to crew health at effective biocide concentrations	Must not require more than 3 years to begin flight demonstration/ Implementation	Must be commercially available or can be made available quickly for evaluation in NASA ground systems	Must not be a Toxic Hazard Level 3 (storage or use)	Must not degrade/ become ineffective under storage conditions (non-use) for up to 3 years
Halogen Binding Resin (Umpqua)	PASS: released 0.5-4 ppm Br over shelf life	PASS: currently at TRL 3 entering Phase II SBIR	PASS: is in NASA-funded testing	PASS	UNKNOWN: but probably if packaged and kept dry
Poly-1-bromo-5-methyl-5 (4'-vinylphenyl) Hydantoin (HaloPure BR)	PASS: 0.1-0.5 ppm Br released	Unknown: Commercial bead filter product, should be largely plug-n-play	PASS	PASS	UNKNOWN: At least 2-year shelf life with desiccant, no data for 3 years
1-bromo- 3-chloro- 5,5-Dimethylhydantoin (BCDMH)	PASS: max 9 ppm	UNKNOWN	PASS	UNKNOWN: Contact dermatitis potential at certain concentrations	PASS: 3 years
1,3-Dibromo-5,5-Dimethylhydantoin (DBDMH)	PASS	UNKNOWN	PASS: Sigma	UNKNOWN	PASS: 3 years
Bromine (Br_2)	Not Evaluated	Not Evaluated	Not Evaluated	FAIL: Toxic gas	Not Evaluated
Bromine Monochloride (BrCl)	Not Evaluated	Not Evaluated	Not Evaluated	FAIL: Toxic gas	Not Evaluated
2,2-Dibromo-3-Nitrilopropionamide	UNKNOWN: Not yet tested for potable water use	UNKNOWN: Requires FDA approval first	PASS: Sigma and Dow/Aquacar	UNKNOWN: Requires FDA approval first	UNKNOWN: but probably, if packaged and kept dry
2-bromo- 2-nitropropane-1,3-diol	UNKNOWN: Not yet tested for potable water use	UNKNOWN: Requires FDA approval first	PASS: Sigma	UNKNOWN: Requires FDA approval first	UNKNOWN: but probably, if packaged and kept dry
Domiphen Bromide	Out of scope				

Option	**Criteria**				
	Must not introduce a known risk to crew health at effective biocide concentrations	Must not require more than 3 years to begin flight demonstration/ implementation	Must be commercially available or can be made available quickly for evaluation in NASA ground systems	Must not be a Toxic Hazard Level 3 (storage or use)	Must not degrade/become ineffective under storage conditions (non-use) for up to 3 years
Sodium dichloroisocyanurate	PASS: 1 ppm	UNKNOWN	PASS: Aquatab	UNKNOWN	PASS: 3-5 years
Di- and Tri-chloroisocyanuric acid	PASS: max 30 ppm	UNKNOWN	PASS: Sigma	UNKNOWN	PASS: Indefinite if stored in cool, dry place
Sodium chlorite (Na(ClO_2))	PASS: 0.7 ppm	UNKNOWN	PASS: Sigma	UNKNOWN	PASS: 3 years
Chloramine-T	PASS: 0.5-2.0 ppm	UNKNOWN	PASS: Sigma	UNKNOWN	PASS: 3 years
Calcium hypochlorite ($Ca(OCl)_2$)	UNKNOWN: Approved for emergency potable water disinfection by the EPA (5 mg/L)	UNKNOWN	PASS: Sigma	UNKNOWN	PASS: 3-5 years
N-Chlorosuccinimide	PASS	UNKNOWN	PASS: Sigma	UNKNOWN	UNKNOWN: probably, structure similar to hydantoin
Chlorine dioxide (ClO_2)	Not Evaluated	Not Evaluated	Not Evaluated	FAIL: Toxic gas	Not Evaluated
Chlorine (Cl_2)	Not Evaluated	Not Evaluated	Not Evaluated	FAIL: Toxic gas	Not Evaluated
Monochloramine(NH_2Cl)	Not Evaluated	Not Evaluated	Not Evaluated	Not Evaluated	FAIL: short shelf life
Sodium hypochlorite (NaOCl)	UNKNOWN: Approved for emergency potable water disinfection by the EPA (~6-8 ppm)	UNKNOWN	PASS: Sigma	UNKNOWN	FAIL: low shelf life at 20 C; pentahydrate form can last 1+ years at 7 C
Halazone	PASS: typical dose 4 ppm	UNKNOWN	PASS: Sigma	UNKNOWN	FAIL: 5-6 months unopened, 3 days upon opening pill bottle

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