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**Endoscope Reprocessing Improvement Suggestion; Is Ozone Water the Answer? By Alvaro Liceaga *MD*; Elvin Mercado *CST*; Lisa Liceaga *M4*; Mary Narcaroti *RNMSHA*. Outpatient Surgery Center. Fullerton, CA**

**Introduction:** The cleanliness and sterility of instruments, is of most importance in the surgical sciences. As we strive to improve our techniques in cleaning, sanitizing and sterilizing equipment, we continue to look for additional agents and techniques. Ozonated water has been shown to be effective in a variety of applications for sanitation. This current test is designed to evaluate cleaning and disinfection of Endoscopes using Cidex®, Ozonated Water and 70% Alcohol Flush. Cidex is a common designation for a variety of solutions used for antimicrobial or disinfection purposes: Cidex OPA, a trade name for a solution with 2.4% Glutaraldehyde as active ingredient. Cidex is universally accepted in the medical / surgical professions as an effective germicide used for antimicrobial or disinfection purposes. Cidex® can be used to disinfect a wide range of medical instruments, made of aluminum, brass, copper, stainless steel, plastics and elastomers. 70% Alcohol Flush is also used an additional chemical sterilant for the high-level disinfection of scopes used in endoscopic procedures.

**Glossary of Terms:**

**Endoscope**: Olympus**EVIS CF-100TL Video Colonoscope with a diameter of 12.9mm and working length of 168cm was used for this study. A colonoscope is a flexible, tubular device using fiber optics to permit visualization and inspection of the colon to look for abnormalities and can also be used to remove or take tissue samples.**

**EndoSwab:** A 3mm absorbent sponge tip with 2.4m wand length to ensure recovery of a representative sample for most endoscope lengths. Clinically clean packaged, the EndoSwab is used to obtain ATP bio load in the main channels of flexible endoscopes that result in biofilm buildup over time. It is compatible with the UltraSnap

**Hygiena’s SystemSURE Luminometer**: is a proprietary device used to measure ATP collected with the swab and results are expressed numerically as Relative Light Units (RLU).

Enzymatic solution: Enzymatic solution contains a synergistic combination of multiple enzymes, which effectively cleans away blood, proteins, fat, carbohydrates, and synthetic liquids from surgical instruments, flexible and rigid scopes, and other general healthcare equipment.

**Luminometer:** Hygiena Luminometer, in conjunction with ATP swabs, use bioluminescence to detect residual ATP as an indicator of surface cleanliness. The presence of ATP on a surface indicates improper cleaning and the presence of contamination, including food residue, allergens and/or bacteria. This implies a potential for the surface to harbor and support bacterial growth.

**Relative Light Units (RLU):** RLU stands for Relative Light Unit and is the unit of measure used in bioluminescence. When a test swab is activated, a bioluminescent reaction occurs, generating light output. Luminometers measure and quantify that light with an RLU output. Because manufacturers use different sensor technologies and algorithms for adding up the photons, RLU measurements will vary from system to system. However, because the ATP bioluminescence reaction is linear, the more ATP present means the more light will be present. This makes comparing systems easy. See (<https://www.hygiena.com/frequent-asked-questions-food-and-beverage.html>).

**Cidex®**: Cidex is a common designation for a variety of solutions used for antimicrobial or disinfection purposes: Cidex OPA, a trade name for a solution with 2.4% Glutaraldehyde as active ingredient. Cidex is universally accepted in the medical / surgical professions as an effective germicide used for antimicrobial or disinfection purposes. Cidex® can be used to disinfect a wide range of medical instruments, made of aluminum, brass, copper, stainless steel, plastics and elastomers.

**Ozonated Water:** Ozonated Water (O3-H20) free radical oxygen molecules react with microbes and effectively kill them via oxidation and degenerate organic compounds. Ozone quickly kills all microorganisms non-selectively, including bacteria, viruses, molds and spores. According to the US EPA Office of Water, ozone is one of the most potent and effective germicides when used in water treatment*.*

**Ozone Properties and O3 Generation**: Ozone is produced using an ozone generating electrical device, an ozone generator, located in the cabinet under the faucet. The ozone gas is dissolved into the water via a venture-type mixing system so that the ozone concentration at the tap outlet remains constant for a given flow rate. A typical faucet used produces a range of ozone dissolved in water at 0.5 to 1.4 ppm. It is generally believed that longer contact time produces greater effectiveness and that at lower concentration fewer O3 overspills go into the atmosphere. In water, ozonated water is produced when gaseous oxygen (O2) molecules are dissociated by an electrical charge into nascent oxygen atoms (O-) which then subsequently collide with oxygen molecules to form the unstable ozone gas molecule (O3) which has very high oxidizing capacity. Water, when “ozonated” (O3-H20), produces free radical oxygen molecules that react with the microbes and effectively kill them via oxidation. Ozone’s antiseptic properties arise from its ability to denigrate cell walls and in some cases cell RNA as well as oxidize organic and non-organic compounds. Ozonated water can effectively kill all microbes and pathogens, including bacteria, viruses, molds and spores. Ozone is fast reacting and is nonselective to all microorganisms. According to the US EPA Office of Water, ozone is one of the most potent and effective germicides when used in water treatment and is effective against bacteria, viruses, mold and protozoan cysts*(US EPA, 1999).* The ozonated water breaks down into oxygen gas and plain pure water in approximately 30 to 75 minutes.

**pH**; A measure of acidity or alkalinity of water soluble substances (pH stands for 'potential of Hydrogen'). A pH value is a number from 1 to 14, with 7 as the middle (neutral) point. Values below 7 indicate acidity which increases as the number decreases, 1 being the most acidic.

**Temperature**: Temperature is usually expressed in degrees**Fahrenheit**or**Celsius**. Zero degrees Celsius is equal to 32 degrees Fahrenheit, and 25 degrees Celsius is equal to 77 degrees Fahrenheit. Ozone concentration can be inversely affected with increased temperature in the water.

**Rinse / Flush**: Rinsing is performed by immersing the device in clean tap water or ozonated water after being soaked enzymatic solution or Cidex solution. It involves washing and wiping the exterior surface of the device. Flushing is then performed by injecting water or air in the internal channels of the device using sterile syringe and tubing adapters to expel remnants of any solution that may be left in the internal channels.

**ATP Monitoring:** ATP monitoring is a rapid testing method used by food and beverage processors to quickly assess the cleanliness of surfaces or liquid samples from such places as CIP systems. Adenosine Triphosphate (ATP) is present in all organic material and is the universal unit of energy used in all living cells. ATP is produced and/or broken down in metabolic processes in all living systems. Processes such as photosynthesis in plants, muscle contraction in humans, respiration in fungi, and fermentation in yeast are all driven by ATP. Therefore, most foods and microbial cells will contain some level of naturally occurring ATP.

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**ATP Testing of Bio-load on Soiled Endoscope:**

We performed a comparison of Cidex® vs. Ozonated Water use following an introduction of organic material such as blood, mucous and saliva in the main biopsy channel of the endoscope device. This test was conducted to determine the effectiveness of Cidex versus Ozone Water on processed (manually cleaned) and high-level disinfection of the endoscope. 70% Alcohol Flush was added to enhance disinfection.

**HYGIENA ATP Cleaning Verification System Tool:** Hygiena’s SystemSURE plus ATP Cleaning Verification System is a tool used to monitor and improve the cleanliness levels of surfaces in healthcare facilities. It uses bioluminescence technology to identify and measure adenosine triphosphate (ATP). ATP is an energy molecule found in all living organisms, including skin, blood, mucus, saliva, viruses, fungi, molds, spores and bacteria. The persistence of ATP on the hand may be an indication of incomplete or improper cleaning technique or a measure of residual ATP from human skin cells. The Hygiena’s SystemSURE Luminometer, is used to measure ATP collected with the swab and results are expressed numerically as Relative Light Units. (RLU).

The temperature, pH and ozone concentration of the ozone water were also obtained.

**Six sets of Endoscope Reprocessing tests were performed: Each received an Enzymatic Soak prior to use of each agent.**

**Set 1: Cidex OPA Soak and Tap Water Rinse**

**Set 2: Ozone Water Soak and Ozone Water Rinse**

**Set 3: Cidex OPA Soak + 70% Alcohol Flush in Endoscope Reprocessing**

**Set 4: Ozone Water Soak + 70% Alcohol Flush in Endoscope Reprocessing**

**Set 5:** **Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush in Endoscope Reprocessing**

**Set 6: Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush in Endoscope Reprocessing**

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**Set 1: Cidex Soak and Tap Water Rinse in Endoscope Reprocessing**

**Method and Procedure:**

The main biopsy channel of the Olympus**EVIS CF-100TL Video Colonoscope** was introduced with combination of blood, mucous and saliva that came from healthy volunteers. Then, an "EndoSwab" that features a foam tip was used to take a sample of the organic material inside the channel. The foam tipped swab with 2.4mm wand length was inserted in the biopsy channel all the way through the distal tip of the device. The swab was taken out from the same channel and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The control number was then established.

**Pre-cleaning at Bed Side**

Pre-cleaning is the first step in reprocessing endoscopes. Pre-cleaning is performed at the bed side immediately after the procedure ends. It is critical to remove all debris and organic material before it has the chance to dry. The exterior of the device was wiped down with clean soft cloth soaked in tap water and the main biopsy channel was flushed with 50 ml of tap water using a syringe.

**Manual Cleaning with Brushing and Flushing Using Regular Tap Water**

After Pre-cleaning was performed, the device was transferred to the Sterile Processing Department for manual cleaning process.   The device was completely submerged in enzymatic solution mixed with regular tap water for a minimum of two minutes according to the manufacturer’s instruction.

During immersion, the main biopsy channel was continuously flushed with the enzymatic solution using 60 ml syringe for 20 seconds. The main biopsy channel was brushed meticulously using special “Endobrushes” designed to fit in the lumen of the device to obtain maximum results. After thorough manual cleaning, regular tap water was introduced to flush the solution out in a separate clean basin for a minimum of 1 minute. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**High Level Disinfection Using Cidex OPA Solution**

Next, using a clean basin, the device was fully submerged in Cidex OPA solution for high-level disinfection. During immersion, a 60 cc syringe was used to continuously inject and flush the device with Cidex OPA solution in the main biopsy channel for 20 seconds. Then, the device was left fully soaked in the Cidex OPA solution for a minimum of 12 minutes according to the manufacturer’s instruction.  The scope was then hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Rinsing Using Regular Tap Water Rinse**

After the device was soaked in the Cidex OPA solution for a minimum of 12 minutes, the main biopsy channel was flushed with air to expel any remnants of Cidex OPA solution inside the channel. The device was then prepared for rinsing using regular tap water. Using a clean basin, the device was fully submerged in regular tap water for rinsing. The main biopsy channel was continuously flushed with tap water for 20 seconds. The device was removed from the basin and transferred to a clean empty basin to remove the remnants of the tap water by flushing the channel with air using a syringe. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Set 1 Results: Cidex OPA Soak and Tap Water Rinse for Endoscope Reprocessing**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Instrument** | **Control**  **RLU** | **Post Enzymatic Soak and Tap Water Rinse**  **RLU** | **Post Cidex Soak and Tap Water Rinse** **RLU**  After 12min soak | **pH/Temperature** | **Ozone Concentration**  **(ppm)** |
| Olympus Colonoscope | **5598** | **7** | **7** | **8.5 / 74F** | **1.15** |
| Olympus Colonoscope | **2035** | **26** | **16** | **8.4 / 74.1F** | **1.13** |
| Average | **3817** | **16.5** | **11.5** | **8.4 / 74 F** | **1.14** |

**Results Set 1: Cidex Soak** **and Tap Water Rinse: 99.69% Average RLU Reduction.**

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**Set 2: Ozonated Water Soak in Endoscope Reprocessing**

**Method and Procedure:**

The main biopsy channel of the Olympus**EVIS CF-100TL Video Colonoscope** was introduced with combination of blood, mucous and saliva that came from healthy volunteers. Then, an "EndoSwab" that features a foam tip was used to take a sample of the organic material inside the channel. The foam tipped swab with 2.4mm wand length was inserted in the biopsy channel all the way through the distal tip of the device. The swab was taken out from the same channel and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The control number was then established.

**Pre-cleaning at Bed Side**

Pre-cleaning is the first step in reprocessing endoscopes. Pre-cleaning is performed at the bed side immediately after the procedure ends. It is critical to remove all debris and organic material before it has the chance to dry. The exterior of the device was wiped down with clean soft cloth soaked in tap water and the main biopsy channel was flushed with 50 ml of tap water using a syringe.

**Manual Cleaning with Brushing and Flushing Using Ozone Water**

After Pre-cleaning was performed, the device was transferred to the Sterile Processing Department for manual cleaning process.   The device was completely submerged in enzymatic solution mixed with ozonated water for a minimum of two minutes according to the manufacturer’s instruction.

During immersion, the main biopsy channel was continuously flushed with the enzymatic solution using 60 ml syringe for 20 seconds. The main biopsy channel was brushed meticulously using special “Endobrushes” designed to fit in the lumen of the device to obtain maximum results. After thorough manual cleaning, ozonated water was introduced to flush the solution out in a separate clean basin for a minimum of 1 minute. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**High Level Disinfection Using Ozonated Water**

Next, using a clean basin, the device was fully submerged in Ozonated Water for high-level disinfection. During immersion, a 60 cc syringe was used to continuously inject and flush the device with ozonated water in the main biopsy channel for 20 seconds. Then, the device was left fully soaked in the ozonated water for a minimum of 12 minutes. (Same recommended contact time for Cidex OPA for comparison).  The scope was then hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Rinsing Using Ozonated Water Rinse**

The device was soaked in the ozonated water for 12 minutes, the main biopsy channel was flushed with air to expel any remnants of water inside the channel. The device was then prepared for rinsing using fresh ozonated water. Using a clean basin, the device was fully submerged in fresh ozonated water for rinsing. The main biopsy channel was continuously flushed with ozonated water for 20 seconds. The device was removed from the basin and transferred to a clean empty basin to remove the remnants of the ozonated water by flushing the channel with air using a syringe. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and read using the Hygiena Luminometer device. The results were then recorded.

**Set 2 Results: Ozonated Water Soak for Endoscope Reprocessing**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Instrument** | **Control** **RLU** | **Post Enzymatic Soak and Ozone Water Rinse**  **RLU** | **Post Ozone Water Soak and Ozone Water Rinse** **RLU**  **12min soak** | **pH/Temperature** | **Ozone Concentration**  **(ppm)** |
| Olympus Colonoscope | **5261** | **13** | **6** | **8.5 / 74F** | **1.12** |
| Olympus Colonoscope | **2348** | **7** | **6** | **8.1 / 73.3 F** | **1.15** |
| Average | **3805** | **10** | **6** | **8.3 / 73.6 F** | **1.14** |

**Set 2 Results: Ozone Water Soak and Ozone Water Rinse = 99.84% Average RLU Reduction.**

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**Set 3: Cidex OPA Soak + 70% Alcohol Flush in Endoscope Reprocessing**

*Note: Set 3 is conducted using the same steps in methods and procedures, pre-cleaning at bed side, manual cleaning and brushing, high-level disinfection and rinsing that were performed for Set 1 (Post Cidex Soak and Tap Water Rinse) with the addition of 70% Isopropyl Alcohol flush as a last step in the reprocessing sequence.*

**70% Isopropyl Alcohol Flush**

To enhance the drying process and help inhibit the growth of microorganisms, 70% Isopropyl alcohol is used for alcohol flushing. Using a 60 cc syringe, 40 cc 70% Isopropyl alcohol is injected into the main biopsy channel, this process was repeated twice. Then, using a syringe, air was injected to the main biopsy channel to expel the remnants of alcohol.  The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Set 3 Results: Cidex OPA Soak + 70% Alcohol Flush in Endoscope Reprocessing**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Instrument** | **Control** | **Post Enzymatic Soak and Tap Water Rinse**  **RLU** | **Post Cidex Soak and Tap Water Rinse** **RLU**  **12min soak** | **70% Isopropyl Alcohol Flush** **RLU:** | **pH/Temperature** | **Ozone Concentration**  **(ppm)** |
| Olympus Colonoscope | **8532** | **8** | **7** | **1** | **8.5 / 74.5F** | **1.12** |

**Set 3 Results: Post Enzymatic Soak and Cidex OPA Soak, Tap Water Rinse + 70% Alcohol Flush = 99.98% RLU Reduction**

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**Set 4: Ozone Water Soak + 70% Alcohol Flush in Endoscope Reprocessing**

*Note: Set 4 is conducted using the same steps in methods and procedures, pre-cleaning at bed side, manual cleaning and brushing, high-level disinfection and rinsing that were performed for Set 2 (Post Ozone Water Soak and Ozone Water Rinse) with the addition of 70% Isopropyl Alcohol flush as a last step in the reprocessing sequence.*

**70% Isopropyl Alcohol Flush**

To enhance the drying process and help inhibit the growth of microorganisms, 70% Isopropyl alcohol is used for alcohol flushing. Using a 60 cc syringe, 40 cc 70% Isopropyl alcohol is injected into the main biopsy channel, this process was repeated twice. Then, using a syringe, air was injected to the main biopsy channel to expel the remnants of alcohol.  The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Set 4 Results: Ozone Water Soak + 70% Alcohol Flush in Endoscope Reprocessing**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Instrument** | **Control** | **Post Enzymatic Soak and Ozone Water**  **RLU Rinse** | **Post Ozone Water Soak and Ozone Water Rinse** **RLU**  **12min soak** | **70% Isopropyl Alcohol Flush** **RLU:** | **pH/Temperature** | **Ozone Concentration**  **(ppm)** |
| Olympus Colonoscope | **7597** | **10** | **7** | **1** | **8.2 / 74.1F** | **1.12** |

**Set 4 Results: Endoscope Reprocessing; Post Enzymatic Soak and Ozone Water Soak** **and Ozone Water Rinse + 70% Alcohol Flush = 99.98% RLU Reduction**

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**Set 5 and 6: Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush in Endoscope Reprocessing (Two Tests Performed)**

*Note: Set 5 and Set 6 were conducted using the same methods and procedures as a confirmatory measure for this specific technique.*

**Method and Procedure:**

The main biopsy channel of the Olympus**EVIS CF-100TL Video Colonoscope** was introduced with combination of blood, mucous and saliva that came from healthy volunteers.

Then, an "EndoSwab" that features a foam tip was used to take a sample of the organic material inside the channel. The foam tipped swab with 2.4mm wand length was inserted in the biopsy channel all the way through the distal tip of the device. The swab was taken out from the same channel and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The control number was then established.

**Pre-cleaning at Bed Side**

Pre-cleaning is the first step in reprocessing endoscopes. Pre-cleaning is performed at the bed side immediately after the procedure ends. It is critical to remove all debris and organic material before it has the chance to dry. The exterior of the device was wiped down with clean soft cloth soaked in tap water and the main biopsy channel was flushed with 50 ml of tap water using a syringe.

**Manual Cleaning with Brushing and Flushing Using Ozone Water**

After Pre-cleaning was performed, the device was transferred to the Sterile Processing Department for manual cleaning process.   The device was completely submerged in enzymatic solution mixed with ozonated water for a minimum of two minutes according to the manufacturer’s instruction.

During immersion, the main biopsy channel was continuously flushed with the enzymatic solution using 60 ml syringe for 20 seconds. The main biopsy channel was brushed meticulously using special “Endobrushes” designed to fit in the lumen of the device to obtain maximum results. After thorough manual cleaning, ozonated water was introduced to flush the solution out in a separate clean basin for a minimum of 1 minute. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**High Level Disinfection Using Cidex OPA Solution**

Next, using a clean basin, the device was fully submerged in Cidex OPA solution for high-level disinfection. During immersion, a 60 cc syringe was used to continuously inject and flush the device with Cidex OPA solution in the main biopsy channel for 20 seconds. Then, the device was left fully soaked in the Cidex OPA solution for a minimum of 12 minutes according to the manufacturer’s instruction.  The scope was then hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a representative sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Rinsing Using Ozonated Water Rinse**

After the device was soaked in the Cidex OPA solution for a minimum of 12 minutes, the main biopsy channel was flushed with air to expel any remnants of solution inside the channel. The device was then prepared for rinsing using fresh ozonated water. Using a clean basin, the device was fully submerged in fresh ozonated water for rinsing. The main biopsy channel was continuously flushed with ozonated water for 20 seconds. The device was removed from the basin and transferred to a clean empty basin to remove the remnants of the ozonated water by flushing the channel with air using a syringe. The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**70% Isopropyl Alcohol Flush**

To enhance the drying process and help inhibit the growth of microorganisms, 70% Isopropyl alcohol is used for alcohol flushing. Using a 60 cc syringe, 40 cc 70% Isopropyl alcohol is injected into the main biopsy channel, this process was repeated twice. Then, using a syringe, air was injected to the main biopsy channel to expel the remnants of alcohol.  The scope was hung to drip and dry for 30 minutes. Then, an EndoSwab was used to obtain a sample and the tip was cut using clean pair of scissors and introduced to the UltraSnap collection device. The liquid-stable reagent was released to mix with the collected sample and then read using the Hygiena Luminometer device. The results were then recorded.

**Set 5 and 6 Results: Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush in Endoscope Reprocessing**

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| --- | --- | --- | --- | --- | --- | --- |
| **Instrument** | **Control** **RLU** | **Post Enzymatic Soak and Ozone Water Rinse** **RLU** | **Post Cidex Soak and Ozone Water Rinse** **RLU**  **12min soak** | **70% Isopropyl Alcohol Flush** **RLU:** | **pH/Temperature** | **Ozone Concentration**  **(ppm)** |
| Olympus Colonoscope  Set 5 | **2741** | **8** | **5** | **1** | **8.5 / 73F** | **1.10** |
| Olympus Colonoscope  Set 6 | **2019** | **10** | **4** | **0** | **8.1 / 73.3 F** | **1.12** |
| Average | **2380** | **13** | **4.5** | **0.5** | **8.3 / 73.1 F** | **1.11** |

**Set 5 and 6 Results: Post Enzymatic Soak and Cidex OPA Soak + Ozone Water Rinse / Flush + 70% Alcohol Flush = 99.98%** **Average RLU Reduction**

**Summary of the Results**

**The Summary of Results in Endoscope Reprocessing are as follows:**

**Set 1: Cidex OPA Soak Percent Reduction of 99.69%**

**Set 2: Ozone Water Soak Percent Reduction of 99.84%**

**Set 3: Cidex OPA Soak + 70% Alcohol Flush Percent Reduction of 99.98%**

**Set 4: Ozone Water Soak + 70% Alcohol Flush Percent Reduction of 99.98%**

**Set 5: Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush Percent Reduction 99.98%**

**Set 6: Cidex OPA Soak + Ozone Water Flush + 70% Alcohol Flush Percent Reduction 99.98%**

**Discussion:**

Generally, Biofilm can corrode endoscopes, instruments and plumbing, cause infections, can adhere to implanted hardware and may not be totally removed during standard processing. Certain instruments such as endoscopes may harbor hidden contaminants due to their inherent design and may be difficult to clean during the disinfection process. Effective processing of sterile endoscope instruments is absolutely required to prevent the spread of pathogens and in reducing the incidence of healthcare associated nosocomial infections, especially with the growing risk of infection with resistant pathogens. Biofilm is present on the instruments after every endoscopic procedure. Biofilm is a complex structure that adheres to surfaces in contact with the endoscope; bacterial biofilm may secrete a slimy protective coating, and are generally resistant to antibiotics and disinfectants.

Factors in effective endoscope cleaning and decontamination are known to include, water quality, cleaning agents used, proper washing method, proper rinsing and drying techniques, staff training practices to prevent personal injury, design of the processing room and maintenance of sterility in storage.

In this study, we saw that the addition of Ozone Water rinses prior to high-level disinfection markedly improved the effectiveness of sanitation and sterilization, especially over Tap Water rinses. We used Ozonated water (O3-H20) generated through a Sanitas® Ozone Faucet System. This was known to possess excellent antimicrobial properties (by ATP reduction), and thus to markedly improve our impression of effectiveness in the sanitation and sterilization process. In other words, Adenosine Tri-Phosphate (ATP) reduction on endoscope instruments was equivalently reduced when ozone water rinsing was added to the regimen. Some additional improvements were achieved when an additional manual 70% Alcohol Flush step was added.

**Conclusion:**

We concluded that the high-level disinfection protocols are overall very effective but may be incomplete depending on agents used. As we strive to improve our disinfection results, we are looking for the addition or substitution of better agents that should result in improvements in high-level disinfection protocol.

In this study, we found that the addition of Ozonated Water into the high-level disinfection protocol made a measured decrease in measured ATP. In fact, we found that substitution of Ozonated Water for Cidex® resulted in equivalent ATP reduction. This alone is a dramatic finding the warrants further evaluation and study.

We must emphasize that RLU of 16 is still considered high after Post Enzymatic and Post Cidex soak with Tap water rinse, perhaps due to recontamination with tap water that is known to contain living organisms.

Further, if ozonated water possesses excellent antimicrobial properties and knowing the toxic nature of Cidex OPA or Glutaraldehyde, perhaps a modification in the endoscope instrument sanitation and sterilization protocol regimen may be indicated. The additional use of Ozone may further help reduce the incidence of endoscope borne nosocomial infection. Additionally, the use of 70% Alcohol Terminal Flush resulted in additional reduction in ATP levels, to 99.98%. This intuitively may be a valuable step to include in standard Endoscope Reprocessing.

**Summary:** Endoscopic Suite Reprocessing Personnel are asked to consider the addition of ozonated water to enhance the sterilization regimen. The use of Ozone Water is recommended by the authors of this report, as it has proven to be of value in reducing ATP levels on the surgical equipment surfaces. This report helps confirm that ozonated water possesses excellent antimicrobial properties and ozone should have a place when added to the existing regimen for a more effective instrument sanitation and sterilization protocol.

Addition of culture testing is recommended to confirm the above hypothesis, perhaps including bacterial, viral, spore and fungal culture testing. Further studies are needed to issue additional recommendations.

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