IslandWater
TECHNOLOGIES

SENTRY-AD™

Real-time Monitoring Of The Impact Of Cleaning Agents On Performance Of Anaerobic Digestion Systems.

1.0 04-2018

SENTRY-AD: Real-time Monitoring Of The Impact Of Cleaning Agents On Performance Of Anaerobic Digestion Systems.

Authors: Colin Ragush, Monica Cella, Jack Ambler, Patrick Kiely

Date: February 15th, 2018.

Executive Summary

SENTRY-AD™ is a real-time, bio-electrode sensor that allows for the monitoring of microbial metabolic activity in the anaerobic digestion (AD) wastewater treatment process. The data generated by the bio-electrode sensor allows operators to monitor the impact of toxic chemicals on microbial health in the AD system. Cleaning agents, biocides, are used for equipment and container sterilization in the food and beverage industry as well as to clean wastewater systems' plumbing. These chemicals will enter the anaerobic digester as part of a waste stream and interact with the sensitive microbial community. Cleaning agents entering the anaerobic digester are suspected to contribute to process instability and failure. Key findings from this study characterizing the impact of cleaning agents on SENTRY-AD output are itemized below:

- **SENTRY-AD** reacts rapidly (<1 min) to cleaning agents and monitors the long-term impact on the microbial community and its recovery.

- Tests with hypochlorite, SpeedConc (contains a Quaternary Ammonium Chloride (QUAT or QAC)) and Peracetic acid demonstrated that **SENTRY-AD** rapidly responds to cleaning agent insertion and captures the extended impact of these agents.

Bench-scale testing has demonstrated that SENTRY-AD has a bright future as a powerful tool for wastewater plant operators to monitor chemical entering the biological treatment process, both scheduled and unscheduled events. The rapid response of the sensor (< 1 min) to a changing water quality environment creates a window of opportunity for operator to act to prevent and / or control the exposure of biology to harmful conditions. Additionally, the data gained has the potential to be instrumental in problem diagnosis and the improvement of operational and management practices.

Introduction

SENTRY-AD, a bio-electrode sensor technology, is a world-first real-time sensor solution that provides direct monitoring of microbial activity of the anaerobic digestion treatment process. The bio-activity data (metabolic activity) of the microbiology is relayed to the operators in real-time, and provides the operators with measurement of the microbial stability - critical for ensuring biological treatment process performance. SENTRY-AD can be integrated with other water quality and operational data to aid in improving and optimizing system performance. There is no measurement delay allowing for immediate notification of process shifts and issues, and the high sampling frequency is instrumental in assessment of: long-term performance, trend and pattern recognition, and optimization.

The probes for this study were submerged directly into the wastewater sample, but could have also be installed inline via a tee junction. In full scale operations, the recommendation is to install multiple probe throughout the process (before, in, and after a digester). The probes are connected to a control panel (with the capacity to host 4 probes), housed in a weatherproof control panel installed nearby. The panel transmits the data to the web server via a wifi or cell connection at a selected interval – typically every minute to allow operators to continually monitor system conditions. The web interface is customized specifically for each installation to provide the customer with real-time diagnostics based on monthly and quarterly review by IWT.

The food and beverage processing industry faces two challenges that are conflicting: (i) sanitization of equipment and containers for food safety and (ii) the disposal of liquid waste high in organic carbon (> 500 mg/l COD). These two challenges conflict because sanitization requires the use of chemical or physical processes to kill off microbes while the treatment of high organic carbon wastewater is most economically achieved by using microbial biological processes that may be adversely impacted by sanitization practices. The presence of chemical sanitization agents in the anaerobic process has been documented to be destabilizing to the process and even result in failure. There is interest from anaerobic digester operators, especially brewers and food processors, in improving monitoring for the controlled addition of waste streams containing sanitizing chemicals and system cleaning agents to improve process stability. The anaerobic digester operators, maintenance staff and, in food and beverage companies, production staff all play important roles in managing cleaning events and accidental discharges that impact the reactor.

SENTRY-AD provides data that facilities can organize around to institute effective management practices and additionally provides a warning when operational practices fail to protect the reactor's biology.
SENTRY-AD tracks in real-time the microbial environment quality by tracking the metabolic activity of the exoelectrogenic species present in a reactor. Exoelectrogenic bacteria utilize simple organic carbon compounds efficiently, such as volatile fatty acids (VFAs) that enter with the effluent or are produced by syntrophic bacteria as a metabolic substrate, and donate electrons onto a metallic electrode. These exoelectrogens are in direct competition with methanogens for VFA substrate. SENTRY-AD rapidly responds to the presence of microbial toxic compounds that affect a species or species in the community responsible for transforming complex organics into volatile fatty acids and then into methane. SENTRY-AD responds to a system's environmental changes, and there is strong evidence that it will respond to operational events such as biofilm preventive maintenance or influent containing sanitizing agents. Potential exists for it to be tool to improve system operation and best practices resulting in increased stability of the anaerobic reactor. The response of SENTRY-AD to three different cleaning agents, sodium hypochorlite, SpeedConc and peracetic acid are presented and discussed.

Materials and Methods

- **1. The SENTRY-AD bench-scale test apparatus was used for testing each cleaning agent. Sensors were pre-inoculated and confirmed for strong and stable sensor response over a period of at least 30 days.**
- **2. The sensor inoculation cup was either fed a complex synthetic feed (with meat extract and casein) or a real wastewater such as brewery or pulp and paper wastewater.**
- **3. The sensor was allowed to achieve a reference baseline, stable response, to compare the impact of a cleaning agent when added to the solution.**
- **4. A known cleaning agent concentration was added into the container with the probe. The signal response was tracked and recorded over time.**

Results and Discussion

When the SENTRY-AD probes were exposed to cleaning agents, the sensor rapidly (< 1 min) responded with an increase of Microbial Electron Transfer (MET). The exact mechanism(s) that lead to the increased MET is under investigation but the data suggests that this is a microbial stress response. As the chemicals attackmicrobial cells, the metabolism of the attacked bacteria increases and/or reserve energy stored as ATP is rapidly utilized and more electrons are given off to the anode. However, cells are damaged and the metabolic activity of all or part of the microbial consortium may be reduced over an extended duration.

4.1 Sodium Hypochlorite

Sodium Hypochlorite, more commonly known as bleach, is a broad-spectrum biocide that attacks the protein of microbes analogously to heating as heat shock protein 33 (Hsp33), a protein that activates to protect other proteins during heating, reacts to the presence of bleach. Akin to overheating, bleach results in denaturing of cell proteins which renders them ineffective, and long term will kill cells and hamper bacterial growth. (Winter et al. 2008)

The response of the SENTRY-AD to sodium hypochlorite at an effective concentration of 3 mg/l (50 mg/l at 6%) is indicative of the broad biocidal impact. Immediately a stress impact is evident with a 50% spike increase of MET (Figure 1). The stress response remains for about 30 minutes, and is followed by a marked decrease in MET as cellular damage becomes apparent in the microbial community and the contact time of the hypochlorite compounds, causing continued cell damage. The impact of the cleaning agent was immediately evident, but the longer-term implications are prolonged as: (i) the SENTRY-AD measurement is contributed to by a diverse community; (ii) the cellular disruption mechanism may not be instantaneous ;(iii) cells may not killed but cell reproduction may be disrupted; (iv) the syntrophic bacteria may be the impacted members and; (v) in these trials the chemical remains present and may still have disinfection efficacy well after addition.

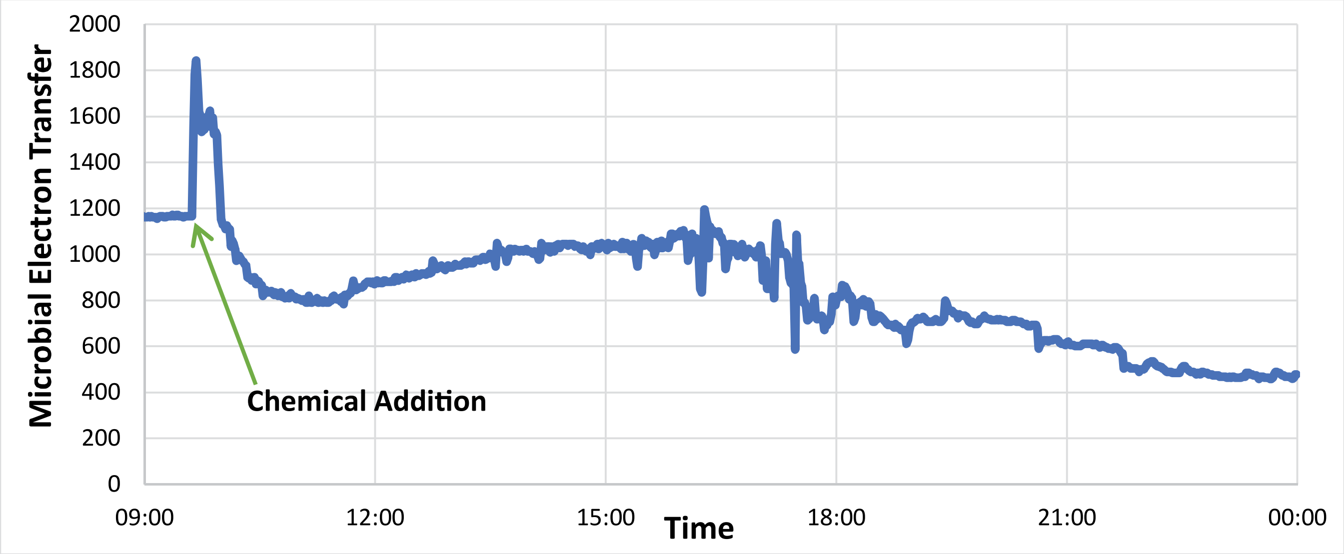

Figure 1. Response of SENTRY-AD to 50 mg/l of 6% commercial sodium hypochlorite.

4.2 SpeedConc (QUATs/ QACs)

SpeedConc is a tradename for a biocide manufactured by Diversey and sold Netherlands. It contains two active ingredients didecyldimethylammonium chloride (DDAC) and amines, N-C12-18-alkyltrimethylendi-, diacetate (10-20% each V/V). DDAC is a Quaternary Ammonium Chloride (QAC) and is commonly used as a biocide. Alkyltrimethylendi is also a known biocide, but the available research on the efficacy and impact of these compounds is limited. QACs disinfection mechanisms are at the cell wall where it will disrupt the lipid bilayer and disrupting intermolecular interactions. There is evidence that QACs are particularly disruptive to methanogens (Tezel 2009; Laopaiboon et al. 2002). The mechanistic reasoning for methanogen sensitivity has not been identified, and it may just be that the methanogens in general are slower to grow and more sensitive.

At the tested concentration of 0.05% V/V (0.5 ml/l) SpeedConc there is no evidence that the exoelectrogenic or syntrophic bacteria were negatively impacted because there was no long-term decrease in MET; however, there is an evident stress response immediately within the community as there is a spike in the MET as demonstrated in Figure 2. The increased MET subsides slowly but is still measurably higher five days after the event. These results would support the findings that methanogens are particularly susceptible to QACs as the increased exoelectrogenic activity is attributed to decreased competition.

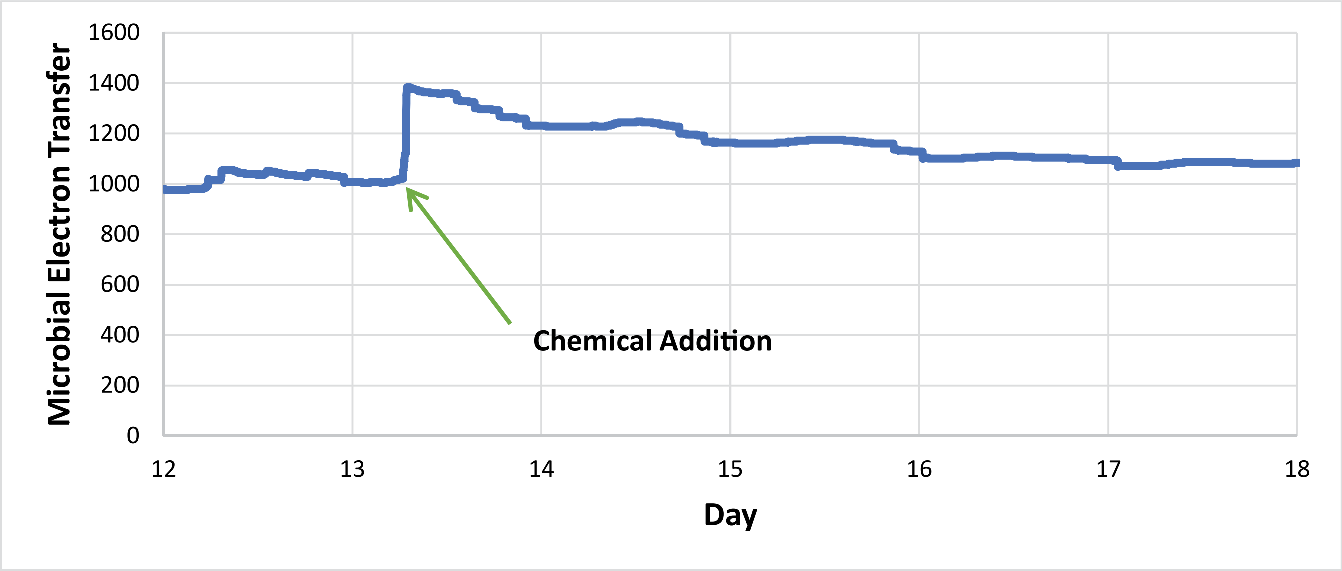

Figure 2. Response of SENTRY-AD to 0.05% V/V of SpeedConc (Quaternary Ammonium Chloride component).

4.3 Peracetic acid

The peracetic acid test was performed as a succession of increasing concentration added daily. Peracetic acid is a common disinfectant in the food and beverage industry and is known to have multiple disinfection mechanism. Additionally, when it decomposes it forms acetic acid. In this test scenario the peracetic acid was delivered in acetic acid (a 39% acetic acid solution). A strong increase in MET was observed, and increased concentration resulted in an increased duration until the MET returned to a steady state as well as a modestly higher peak MET (Figure 3). The increase is likely partially attributed to the addition of acetic acid, a VFA that exoelectrogens are effective at utilizing. However, the concentrations added are small in the context of the elicited response. To our knowledge, the biofilm was stable prior to the test and additional VFAs would not have a major impact unless the biofilm was substrate limited, which there was no evidence of in this test. A component of the feed was 250 mg/l sodium acetate (182 mg/l acetate) which is substantially greater than what was added with the cleaning agent (approximately 2.5 mg/l acetic acid and 2.5 mg/l peracetic acid), and the response to the addition of the peracetic acid was asymmetric.

The voltage in this test was intended to be stable, however as Figure 3 shows the voltage response oscillated inversely to MET. This likely was a circuit limitation in this iteration of design. Figure 4 is a graph of Electron Transfer Resistance with the outliers, caused by circuit instability removed. This is not resistance in the traditional circuit sense, but can be conceptualized as a representation of the ease with which exoelectrogens can donate electrons to the anode. This measure will be dependent upon the substrate concentration, bulk fluid characteristics, anode/cathode properties and biofilm. With successive additions of peracetic acid there is a diminishing Electron Transfer Resistance at the stabilized METC. This is occurring with diminishing decrease in change despite increasing addition and would suggest that the biofilm has been altered in such a way that benefit the exoelectrogenic bacteria. Further, if substrate limitation was material to this test scenario the Electron Transfer Resistance would decrease as substrate limitation imposes a limitation on MET.

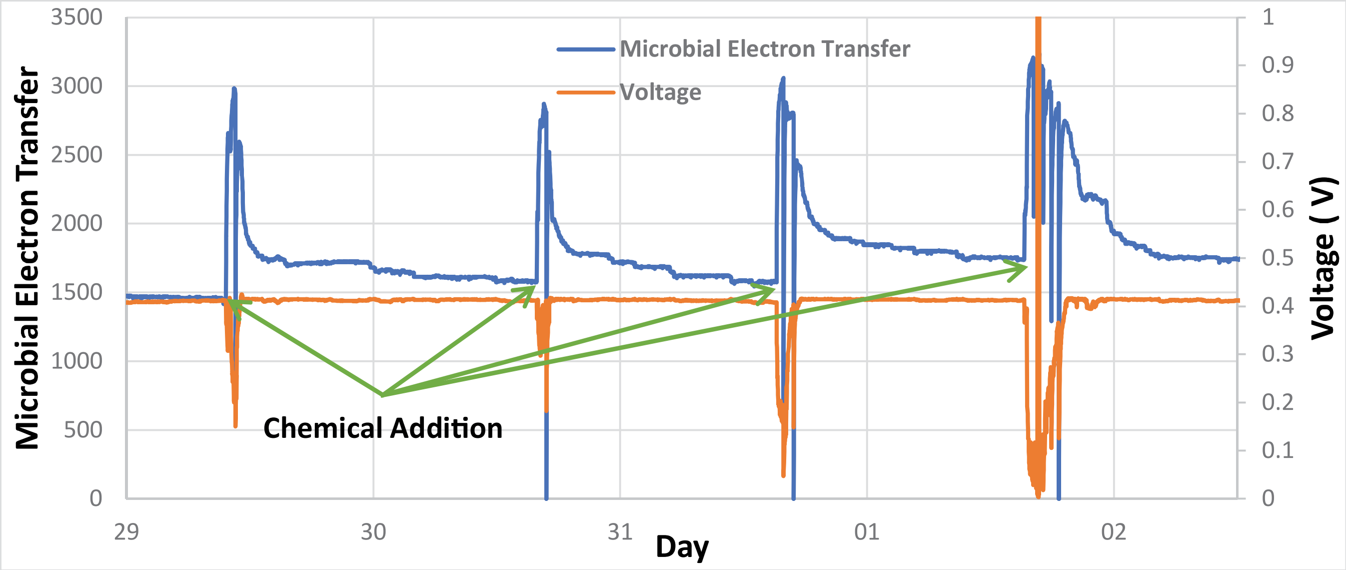

Figure 3. Response of SENTRY-AD to Peracetic acid. In succession 5,10,20, and 50 mg/l.

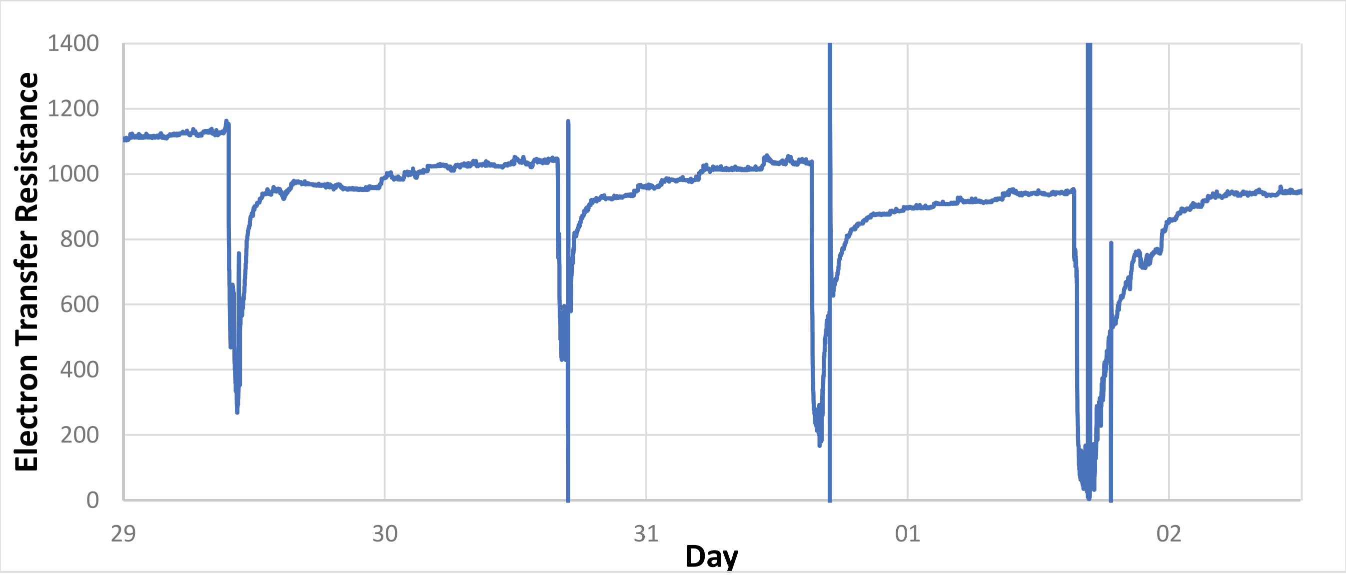

Figure 4 Response of Sentry-AD to Peracetic acid. In succession 5, 10, 20, and 50 mg/l. Scaled voltage/microbial electron transfer provides a representation of the electrical resistance exoelectrogenic bacteria are experiencing when donating electrons to the anode. This 'resistance' would be expected to be stable under mature biofilm conditions with stable feeding. Outliers caused by extremely low MET were removed.

Conclusion

A stress response, identified as a rapid increase in MET, was recorded with all three cleaning agents and acted as a strong warning of a biologically toxic chemical entering the vessel. The long term impact on sensor response was more variable and could be an increase or decrease from the previously stable MET. The long term impact is more variable because there are multiple disinfection mechanisms and the impact on microbes are likely species dependent.

In the case of sodium hypochlorite, the whole community was disrupted and resulted in a continued weakened response. While in the cases of SpeedConc and Peracetic acid it appears that exoelectrogenic species were not strongly impacted, but other microbial species were. SpeedConc resulted in a lasting increase in MET production, and evidence supports the belief that the methanogenic populations are particularly sensitive to this chemical compound. Our hypothesis is that the exoelectrogenic community is reacting to VFA substrate being present in solution as a result of the selective inhibition of methanogenic populations. Peracetic acid had a similar positive impact on MET, whereby exoelectrogenic bacteria were not impacted, but the baseline MET of the sensor increased with every subsequent addition of peracetic acid. The increased baseline is evidence of long-term impact to microbial community, and that likely the impact was on microbes competing for substrate with exoelectrogens. Alternatively, there may be another mechanism responsible (ex. selection) either within the exolectrogens' or syntrophic bacteria's populations.

Bench scale testing has demonstrated the potential of SENTRY-AD to rapidly identify the insertion of a cleaning agent into an anaerobic digester, as well as its ability to track the long term impact of the cleaning agent on microbial performance. The instaneous response provides operators the opportunity to avoid, diagnose, and mitigate the impact of toxic chemicals entering the biologically sensitive anaerobic digester and provides novel data to aid in plant cleaning and system loading management. The microbial community innoculated on the probe is robust enough that changes can be indentified without inactivating the probe. This robustness means that SENTRY-AD not only can identify an event, but can remain in-situ to provide guidance to operators as they work to return the process back to stable and optimal operation.

References

Laopaiboon, L., Hall, S., & Smith, R. (2002). The Eeffect of a quaternary ammonium biocide on the performance and characteristics of laboratory-scale rotating biological contactors. *Journal of Applied Microbiology, 93(6)*, 1051-1058.

Tezel, U. (2009). *Fate and Effect of Quaternary Ammonium Compounds in Bological Systems: Thesis.* Georgia: Georgia Institute of Technology.

Winter, J., Ilbert, M., Graf, P., Ozcelik, D., & Jakob, U. (2008). Bleach activates a redox-regulated chaperone by oxidative protein unfolding. *Cell, 135(*4), 691-701. doi:10.1016/j.cell.2008.09.024

Contact

www.SentryWaterTech.com
www.islandWaterTech.com

info@IslandWaterTech.com
(+1) 902 361-2085

Island Water Technologies Inc
65 Watts Avenue, Unit 9
Charlottetown, PE, C1E 2B7
Canada