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A systematic literature review of sanitizer efficacy to remove *Listeria monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *Escherichia coli* biofilms from food processing surfaces

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A systematic literature review of sanitizer efficacy to remove *Listeria monocytogenes*,
Salmonella spp., and Shiga toxin-producing *Escherichia coli* biofilms from food processing
surfaces

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Spring 2021

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Abstract

Sanitizing in food production environments is essential to prevent, reduce, and/or eliminate foodborne pathogens. Biofilms consist of one or more different types of microorganisms and can grow on numerous types of surfaces (Costerton,1999). SLRs provide transparency about what steps were taken to acquire the sources included in the analysis (Liberati et al.,2009; Moher et al., 2009). The references obtained from the databases were based on specific eligibility criteria to ensure reproducible results. The inclusion criteria included six surface types (stainless steel, glass, plastic, polyurethane, PVC, rubber), seven sanitizer types (anionic acid, benzalkonium chloride, iodine, iodophor, peracetic acid, quaternary ammonium, and sodium hypochlorite), three bacteria types (*L. monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *E. coli*), biofilm methodology (including time, temperature, and media), starting concentration and ending concentration or log reductions present, units in log CFU/cm², stating whether the biofilm was single species or multi species, sanitizer concentration, sanitizer contact time, temperature of sanitizer application, neutralizer used, and biofilm preparation. The outcomes from this SLR will help fill knowledge gaps for future biofilm research and improve biofilm removal with chemical sanitizers. Overall, this study brought to light many future topics of research as well as issues with biofilm removal that can be improved from past research.

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Introduction

It is estimated that annually in the United States, one in six persons contract a foodborne illness (Scallan et al., 2011). Food can become contaminated when pathogens transfer from the food production environment to the food product (Zhao et al. 2017). Sanitizing in food production environments is essential to prevent, reduce, and/or eliminate foodborne pathogens. Biofilms consist of one or more different types of microorganisms and can grow on numerous types of surfaces (Costerton, 1999). Cell attachment in biofilms is initiated when microorganisms are attracted to organic molecules from food that were deposited on the surface. Colonies form as a small number of bacteria cells and then grow into larger groups. Biofilm formation is a complex process. Briefly hydrated extracellular polymeric substances (EPS) containing polysaccharides, proteins, phospholipids, teichoic acid, and nucleic acids form a sessile environment on a given surface. This sessile environment can be used to sustain and protect microorganisms from harsh environments in the absence of additional nutrient supplements (Flemming & Wingender, 2010). Once formed, EPS disruption and biofilm removal is difficult and often leads to a reduced efficacy of cleaning and sanitizing practices in the food industry (Shi & Zhu, 2009; Flemming & Wingender, 2010).

Biofilms are very difficult to remove from surfaces. Bacterial cells can be dispersed from biofilms as part of the biofilm formation cycle and can result in a recurring source of contamination in a food production environment (Zhao et al, 2017). Having a surface with a biofilm can lead to product contamination as well as additional sites of biofilm formation throughout a facility, which is a major concern within the food industry (Alvarez-Ordóñez et al.,

2019). The ability to remove biofilms can be influenced by microorganism types, surface type, and temperature, among other factors (Shi & Zhu, 2009; Phillips, 2016). *Listeria monocytogenes*, *Salmonella enterica*, and Shiga toxin-producing *Escherichia coli* (STEC) three major foodborne pathogens have been studied by researchers to better understand their fate within biofilms following sanitization treatments (Pan et al., 2006). Cleaning and sanitizing are crucial steps to remove biofilms, yet a collective body of literature evaluating biofilm formation methodology, surface type, and sanitizer (type, concentration, contact time) has not been developed. It is important to be able to compare sanitizer efficacy for major foodborne pathogens in the food industry and move towards a standardized approach to biofilm removal.

Systematic literature reviews (SLRs) are an important resource within the scientific field that provide information on a selected topic from specific sources and criteria. SLRs provide transparency about what steps were taken to acquire the sources included in the analysis (Liberati et al., 2009; Moher et al., 2009). Systematic literature reviews advance research by focusing on quality studies and identification of weaknesses so these studies can be excluded from the analysis. These SLRs are performed by: 1) finding published knowledge generated on a specific topic; 2) including relevant studies that fit the scope of a particular topic; and 3) questioning and excluding studies that do not meet inclusion criteria. The inclusion criteria for this SLR were determined by investigating peer-reviewed publications focused on biofilm removal with chemical sanitizers. The inclusion criteria included six surface types (stainless steel, glass, plastic, polyurethane, PVC, rubber), seven sanitizer types (anionic acid, benzalkonium chloride, iodine, iodophor, peracetic acid, quaternary ammonium, and sodium hypochlorite), three bacteria types (*L. monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *E. coli*), biofilm methodology (including time, temperature, and media), starting concentration and ending

concentration or log reductions present, units in log CFU/cm², stating whether the biofilm was single species or multi species, sanitizer concentration, sanitizer contact time, temperature of sanitizer application, neutralizer used, and biofilm preparation. These criteria were developed based on reviewer past knowledge of the food industry as well as preliminary review of the literature regarding the most common surfaces, sanitizers, foodborne pathogens, and biofilm methodology practices.

The goal of this SLR is to determine the efficacy of sanitizers for the removal of *L. monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *E. coli* in single and multispecies biofilms from food processing surfaces as influenced by biofilm formation methodology. The first objective was to identify peer-reviewed publications from relevant databases based on inclusion/eligibility criteria and key terms that fit the scope of the SLR. The second objective was to screen extracted citations based on journal article title/abstract and to extract data from the peer-reviewed literature that fits the scope of the SLR. The final objective was to compile and summarize extracted data in text, tables, and figures that can be easily interpreted by researchers and food industry personnel.

Methods

The databases selected for this SLR were based on suggestions from the University of Arkansas librarians, specifically in the medical and agricultural science fields. The databases that were used for this SLR are CAB Abstracts (Ebsco), Food Science and Technology Abstracts (FSTA), ScienceDirect, AGRICOLA, and Web of Science.

The references obtained from the databases were based on specific eligibility criteria to ensure reproducible results. The criteria are as follows: language – English; publication period – January 1980–October 2020; geographical area – world; publication type – peer-reviewed. The search keywords for this SLR are listed in Table 1.

The eligibility criteria included only studies investigating *L. monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *E. coli* biofilms. Additionally, only studies investigating biofilm removal from surface materials were included. Studies that did not directly quantify the concentration of bacteria removed per surface area were excluded unless an indirect method was validated via cell counting method (flow cytometry, standard plate counts). Additionally, studies that did not state the starting concentration of biofilms (control) were not included in the analysis. Microscopy imaging studies were excluded if it is unclear whether researchers randomly selected images or if a consistent image location selection among multiple samples was not explicitly stated (Wilson et al., 2017). If sanitizer application temperature was reported as room temperature, or if no temperature was provided, a temperature of 22°C was designated for each study. Studies that reported percent (%) concentration of chemicals used for biofilm removal were converted to parts per million (ppm) for comparison across all studies by multiplying the % concentration by 10,000. Chemical concentrations above 400 ppm were excluded from the SLR as sanitizer concentrations typically range from 200 to 400 ppm in the

food industry, and higher concentrations may leave residual sanitizer on the production surface. This could then transfer to the food product causing it to be contaminated. The data were extracted by two independent reviewers in a Microsoft Excel document where data were compared between the reviewers.

Results

Initially, there were 1786 articles that were identified for the study. Sources came from five databases and each supplied a different number of sources: 140 from Agricola, 149 from CAB Abstract Archives, 132 from Food Science and Technology Abstract, 977 from Science Direct, and 388 from Web of Science (Figure 1). After the initial search, 428 duplicate sources were removed. The study proceeded to the title screening with 1358 sources. There were 1146 irrelevant papers removed from the second screening because these sources did not meet the criteria for the study. Some of these studies looked at excluded bacterial types, incorrect surface types, incorrect sanitizer types, etc. Once abstracts were screened for inclusion, there were 131 records identified for full-text screening. There were 81 records removed during the abstract screening. The records were removed for incorrect bacteria, surface types, biofilm methodology, etc. At this point, peer-reviewed references from the last five years were included in the analysis (n=51). During the full text screening from 2016-2020, 40 irrelevant records were removed. Records were removed primarily based on irrelevant study designs (36 references), which was based on a lack of starting concentration $\log \text{CFU}/\text{cm}^2$ provided by authors, chemical concentrations above 300 ppm, or other experimental designs out of scope of this paper (Figure 1). The different criteria above was decided upon throughout the process of the SLR. Not having a starting concentration would have made it impossible to quantify the log reductions. The

chemical concentrations above 300 ppm are not typically found in the industry, so this is why these concentrations were excluded. The studies also needed to be laid out the same including biofilm formation, methodology, and removal. Each of these experimental designs are very important to have to allow the studies to be comparable. The data from the studies included in this SLR were extracted and placed into Microsoft Excel spreadsheets. The data for each reference were placed in separate tabs.

Within this SLR, there were three bacteria types which included *L. monocytogenes*, *Salmonella* spp., and Shiga toxin-producing *E. coli*. These bacteria were chosen as they are well known in the food industry to be problematic. *Salmonella* spp. were researched in most of the studies included. Of the eleven studies included in the SLR, 9 used *Salmonella* spp., 4 used *L. monocytogenes* and 2 used STEC.

Biofilm formation methodology was also taken into account for this SLR. The formation of biofilms varied for each study. For example, Kim et al. (2016) formed biofilms under 24-hour incubation periods at 30° C using tryptic soy broth. Meanwhile, Kumawaza et al. (2016) formed two biofilms under two conditions. The first biofilm was formed with during a 48-hour incubation at 22°C using Tryptose phosphate broth, and the second biofilm was formed over a 48-hour incubation at 22° C using Peptone glucose phosphate broth. These are three very differently formed biofilms yet are all included within this SLR as they each fit the criteria.

There were seven types of sanitizers included in this SLR. The sanitizers are as follows: anionic acid, benzalkonium chloride, iodine, iodophor, peracetic acid, quaternary ammonium, and sodium hypochlorite. With the seven sanitizers included in the SLR, there were 389 data points. These data points are individual pieces of data that were extracted from each paper (n=11) and placed into the Excel document. Ban et al. (2016) had 108 data points while Pang et

al. (2017) only had 24 data points. Of the seven types of sanitizers in these studies, sodium hypochlorite and quaternary ammonium were the most common with 175 and 128 data points, respectively, which together represented (303/389) 78% of the studies.

Each study used different concentrations and contact times for the sanitizers (Table 2). Each study combined different concentrations with different contact times. The median sanitizer concentration was 60 ppm, and the mean was 103.4 ppm. The sanitizer concentrations evaluated ranged from 10 to 300 ppm. The median sanitizer contact time was 5 minutes, and the mean was 15.5 minutes. The sanitizer contact time ranged from 30 seconds to 360 minutes.

Of the six surface types, stainless steel was the predominate surface investigated for biofilm removal with chemical sanitizers. Stainless steel was investigated in 300 of the data points, while glass, plastic, polyurethane, PVC, and rubber made up the other 88 data points. The second most common was PVC which made up 30 data points, and the third most common was rubber with 24 data points. Plastic, glass, and polyurethane comprised the remaining 34 data points and had 4, 15, and 15 data points, respectively.

Discussion

Systematic literature reviews are important resources within the scientific field. This type of research provides information on a variety of topics which come from specific sources and have set criteria. SLRs allow for past experiments to be compared to one another to see the best way for the topic to be done. SLRs are also a way to collate knowledge generated on a topic which can be used for decision making, training purposes, and identification in research gaps. Systematic literature reviews add so much to the scientific community.

In this case, biofilms are pesky issues within the food industry. Biofilms are very complex in formation. Biofilms form when EPS are deposited on a surface and creates an ideal environment for microorganisms to grow upon as there are plenty of nutrients to sustain life. Many types of microorganisms can live within biofilms, but typically bacterial cells are found dispersed throughout. Once a biofilm is created, it is very difficult to remove so it is critical that appropriate sanitation practices are available for biofilm removal. To determine best practices, research must be conducted. Therefore, this SLR looks at many studies to compare sanitation processes for biofilm removal.

Given the time period allocated for this SLR, there were some issues with completion combined with gaps within the data. Due to COVID-19 and the need to adjust from a lab-based project, the SLR was started in October 2020 and was required to be completed by April 1, 2021 as was an honor thesis project for a graduating senior. Although the time period given was quite short, numerous hours were spent on this project. Data will continue to be analyzed in 2021 to compare results among the included peer-reviewed sources despite different approaches. Each study has different parameters for the experiment and was written and communicated differently. For example, Shi et al. (2016) had 30 seconds of sanitizer contact time with a 200 or 200,000 ppm Peracetic acid (PAA) or NaOCl concentration, while Pang et al. (2020) reported a 15-minute contact time at sanitizer concentration of 200 ppm Quaternary Ammonium (QAC) concentration. If both of these studies had the same sanitizer and the same sanitizer concentration, then these results could be directly compared when looking at the log reduction values. Yet, both of these studies use different sanitizer contact times and sanitizers. Unfortunately, few studies were consistent which limits the comparisons among studies. This

will definitely be something that will need to be further looked into for this SLR. Determining how to fully analyze the data will allow this study to be completed.

There are usually multiple steps to a sanitation program. It begins with cleaning which is essentially removing debris. After cleaning the surface, a sanitizer is applied. A sanitizer reduces the number of microorganisms on a surface to a safe level for the general population (CDC, 2021). Following a sanitization step, a disinfectant, designed to kill microorganisms, may be applied.

Something that is missing from the current literature on our selected research topic is cohesiveness across experimental designs. The studies all have different variables which yields different data. Since each paper is not cohesive it makes it challenging to analyze the data across studies. Next steps for this SLR include determining how to analyze the data. These can be generally compared by sanitizer concentration, sanitizer contact time, surface type, and bacteria type. Each study had different recovery methods of the biofilms, the temperature in which the sanitizer was applied, and biofilm preparations. Recovery methods varied by either sonication or glass beads. The temperatures in which the sanitizer was applied was typically between 22-25°C. There were few outliers, but differences between studies made it difficult to compare them.

The different bacteria strains make comparisons within this SLR difficult, especially with *L. monocytogenes*, *Salmonella*, and STEC on different surfaces with separate sanitizers yielding various log reductions. Wang et al. (2020) specifically looked at *E. coli* O157:H7 strains 110, 141, 144, 168 and 170 and *Salmonella* serovars including *S. Dublin* strain 519, *S. Anatum* strain 574, *S. Montevideo* strain 570, *S. Newport* strain 534, and *S. Typhimurium* strain 554. Pang et al. (2017) investigated *S. Typhimurium* CDC 6516-60 and *S. Enteritidis* CDC K-1891. Comparing the log reduction of different bacterial species is challenging and should be carefully interpreted.

Each bacterial type, as well as individual strains between bacterial type makes comparing individual studies difficult as the differences between the strains within a genus may be of importance with respect to the results of this SLR. For example, Dong et al. (2003) observed differences in the survival of *Salmonella* serovars on alfalfa sprouts. Each of the *Salmonella* serovars had high colonization number inside of the seedlings, yet the way each serovar infected the seedlings was different. The serovars infected the seedlings at different locations. Some infected at the endophytic location while others infected the rhizosphere. Since serovars may respond differently as seen by Dong et al. (2003), this should be considered when interpreting results, and future studies may need to evaluate multiple serovars for sanitizer use.

The main type of bacteria used in the 11 studies was *Salmonella*. *Salmonella* Typhimurium and Enteritidis were the most common *Salmonella* serovars evaluated. There were eight studies that investigated *S. Typhimurium* and seven that investigated *S. Enteritidis*. Of the bacteria, only *S. Typhimurium* CDC 6516-60 was used in multiple studies. Three different studies, Sarjit et al. (2016), Shi et al. (2016), and Pang et al. (2017), used *S. Typhimurium* CDC 6516-60 for their investigations. There were no similarities among any of the *L. monocytogenes* or STEC strains used in different studies.

There were ten different sanitizer contact times and ten sanitizer concentrations in the eleven studies included. There was limited overlap, as many studies had different contact times and different sanitizer concentrations. Pang et al. (2018) and Pang et al. (2020) both used 200ppm QAC sanitizer, yet they differed on their contact times. Pang et al. (2018) used 5, 10, and 15 minutes of contact time while Pang et al. (2020) used 10, 60, and 360 minutes of contact time. The many differences in contact times and sanitizer concentrations made it difficult to create tables that directly compared the studies.

Surface type was another added complexity to this research. Each surface type reacts to sanitizers differently. Depending on the surface type and the sanitizer, the bacterial log reduction was different. There is no standard that can be implemented for all types of surfaces. With that said, there were six different surface types. Stainless steel made up the majority as it is the most common industry surface, so the most research has been conducted on this surface type. PVC and rubber were the other two most common surfaces. This of course makes sense since these surfaces are also commonly found within the industry.

For the future, studies should focus on the sanitizer concentration. This would either confirm that the current concentration kills enough bacteria without risking the safety for human consumption or will determine that the concentration is too low or too high. Some studies including Gkana et al. (2017) and Ban et al. (2016) used very low concentrations of sanitizer. The concentrations were 10 and 50 ppm of NaOCl, PAA, and BZK for Gkana et al. and 20, 50, and 100 ppm of Benzalkonium chloride (BZK), H₂O₂, Iodophor, and NaOCl respectively. These concentrations may be too low to inactivate bacterial biofilms. On the other hand, Sarjit et al. (2017) and Wang et al. (2020) used much higher sanitizer concentrations. The concentrations were 40, 50, and 60 ppm of NaOCl and 40, 50, and 60 ppm of Trisodium Phosphate Anhydrous for Sarjit et al. (2017) and 300 ppm of QAC for Wang et al. (2020). Overall, all of the biofilms were reduced in numbers, but some were more effective than others. In the food industry, 200-400 ppm is typically the maximum concentration (FDA, 2020) allowed as higher concentrations may be unsafe to consumers if not followed by a rinse step. There can be residual sanitizer left on the production surface which could transfer to the food product. This is what can become an issue if the sanitizer concentration is too high. Several studies were removed from this SLR as

the concentration of the sanitizers used were at disinfectant levels, which did not fit the inclusion criteria of this study.

Another item that needs to be studied is the sanitizer contact time. Shi et al. (2016) only applied a sanitizer contact time of 30 seconds with 200 ppm sanitizer concentrations. The log reduction for Shi et al. (2016) for this contact time ranged from 1.41-3.35 log CFU/cm². This even with a high sanitizer concentration may be too short of duration to fully sanitize a surface. Wang et al. (2020) uses 360 minutes to sanitize the surfaces in their study with 300 ppm sanitizer concentrations. The log reduction for this contact time ranged from 4.25-6.49 log CFU/cm². Although this amount of time does sanitize the surface the best, it is not practical for the food industry. In a food production sense, there is not a ton of time to deal with sanitizing surfaces. Something that is long enough to kill microorganisms without taking away prime food production time is key to a future study (CFR, 2020). Depending on the type of bacteria and sanitizer concentration, the contact time may need to be adjusted. Additionally, researchers should collaborate with industry personnel to determine feasible contact times for food processing operations.

Throughout the 11 studies, NaOCl and QAC were the most commonly studied sanitizers. With that being said, QAC achieved the greatest bacterial log reductions. Pang et al. (2018), Wang et al. (2020), Sarjit et al. (2016) and Pang et al. (2020) all used QAC and had high bacterial log reductions. Pang et al. (2018), Sarjit et al. (2016), Wang et al. (2020), and Pang et al. (2020) all had bacterial log reductions of 6 log CFU/ cm² or higher. Overall, QAC was a very effective sanitizer. The least effective sanitizer was iodine. Iodine was only used in one study, Kumawaza et al. (2016). In this study the largest bacterial log reduction was 0.1 log CFU/cm².

The biofilm forming methodology was generally consistent. Most used tryptic soy broth and incubated the biofilms for 24-48 hours between 20-30° C. There were two outliers including Kumawaza et al. (2016) and Wang et al. (2020). Kumawaza et al. (2016) used tryptose phosphate broth and peptone glucose phosphate broth incubated for 48 hours while Wang et al. (2020) used Lennox broth without salt for 72 hours. Pang et al. (2018) had the longest incubation time was 336 hours, and the shortest incubation time was 24 hours which was used for multiple studies. For future studies, tryptic soy broth incubated for 24-48 hours between 20-30° C should be researched as these parameters were most commonly used.

Conclusion

Systematic literature reviews are a very important resource as they help to improve the scientific research community. SLRs compile records that fit explicit inclusion criteria for analysis. This allows for scientists to develop and investigate more optimal approaches and fill gaps to research questions that remain to be tested. This SLR was performed to determine the efficacy of chemical sanitizers on the removal of *L. monocytogenes*, *Salmonella*, and STEC biofilms from food processing surfaces based on experimental design and many other factors. Outcomes from this SLR will help fill knowledge gaps for future biofilm research and improve biofilm removal with chemical sanitizers. Overall, this study brought to light many future topics of research as well as issues with biofilm removal that can be improved from past research.

References

- Alvarez-Ordóñez, A., Coughlan, L.M., Braindet, R., Cotter, P.D. (2019). Biofilms in food processing environments: Challenges and opportunities. *Annu. Rev. Food Sci. Technol.* 10, 173-195.
- Ban, G., & Kang, D. (2016). Effect of sanitizer combined with steam heating on the inactivation of foodborne pathogens in a biofilm on stainless steel. *Food Microbiol.* 55, 47-54.
- Center for Disease Control and Prevention. (2021). The Regulatory Framework for Disinfectants and Sterilants. <https://www.cdc.gov/infectioncontrol/guidelines/disinfection/disinfection-methods/regulatory-framework.html>.
- CFR - Code of Federal Regulations Title 21. (2020). Retrieved from <https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/cfrsearch.cfm?fr=178.1010>
- <https://www.cdc.gov/coronavirus/2019-ncov/community/schools-childcare/clean-disinfect-hygiene.html>
- Costerton, J.W. (1999). Introduction to biofilm. *Int. J. Antimicrob. Agents* 11, 217-221.
- Dong, Yuemei, et al. “Kinetics and Strain Specificity of Rhizosphere and Endophytic Colonization by Enteric Bacteria on Seedlings of *Medicago Sativa* and *Medicago Truncatula*.” *Applied and Environmental Microbiology*, vol. 69, no. 3, 2003.
- Flemming, H., & Wingender, J. (2010). The biofilm matrix. *Nat. Rev. Microbiol.* 8, 623-633.
- Gkana, E. N., Giaouris, E. D., Doulgeraki, A. I., Kathariou, S. E., & Nychas, G. U. (2017). Biofilm formation by *Salmonella* Typhimurium and *Staphylococcus aureus* on stainless steel under either mono- or dual-species multi-strain conditions and resistance of sessile communities to sub-lethal chemical disinfection. *Food Control* 73, 838-846.
- Kim, M., Park, S. Y., & Ha, S. (2016). Synergistic effect of a combination of ultraviolet–C irradiation and sodium hypochlorite to reduce *Listeria monocytogenes* biofilms on stainless steel and eggshell surfaces. *Food Control* 70, 103-109.
- Liberati, A., Altman, D.G., Tetzlaff, J., Mulrow, C., Gøtzche, P.C., Ioannidis, J.P.A., Clarke, M., Devereaux, P.J., Kleijnen, J., Moher, D. (2009). The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: Explanation and elaboration. *PLoS Med.* 6(7), e1000100.
- Moher, D., Liberati, A., Tetzlaff, J., Altman, D.G. (2009). Preferred reporting items for systematic reviews and meta-analysis: The PRISMA statement. *PLoS Med.* 6(7), e1000097.
- Pan, Y., Breidt, Jr., F., Kathariou, S. (2006). Resistance of *Listeria monocytogenes* biofilms to sanitizing agents in a simulated food processing environment. *Appl. Environ. Microbiol.* 72(12), 7711-7717.

- Pang, X., Yang, Y., & Yuk, H. (2017). Biofilm formation and disinfectant resistance of *Salmonella* spp. in mono- and dual-species with *Pseudomonas aeruginosa*. *J Appl. Microbiol.* 123(3), 651-660.
- Pang, X., Wong, C., Chung, H., & Yuk, H. (2019). Biofilm formation of *Listeria monocytogenes* and its resistance to quaternary ammonium compounds in a simulated salmon processing environment. *Food Control* 98, 200-208.
- Pang, X., Chen, L., & Yuk, H. (2020). Stress response and survival of *Salmonella* Enteritidis in single and dual species biofilms with *Pseudomonas fluorescens* following repeated exposure to quaternary ammonium compounds. *Int. J. Food Microbiol.* 325, 108643.
- Phillips, C.A. (2016). Bacterial biofilms in food processing environments: A review of recent developments in chemical and biological control. *Int. J. Food Sci. Technol.* 51, 1731-1743.
- Ronner, A. B., & Wong, A. C. (1993). Biofilm development and sanitizer inactivation of *Listeria monocytogenes* and *Salmonella typhimurium* on stainless steel and Buna-n rubber. *J. Food Protect.* 56(9), 750-758.
- Sarjit, A., & Dykes, G. A. (2017). Antimicrobial activity of trisodium phosphate and sodium hypochlorite against *Salmonella* biofilms on abiotic surfaces with and without soiling with chicken juice. *Food Control* 73, 1016-1022.
- Scallan, E., Hoekstra, R.M., Angulo, F.J., Tauxe, R.V., Widdowson, M.A., Roy, S.L., Jones, J.L., Griffin, P.M. (2011). Foodborne illness acquired in the United States—Major pathogens. *Emerg. Infect. Dis.* 17, 7-15.
- Shi, X., Zhu, X. (2009). Biofilm formation and food safety in food industries. *Trends Food Sci. Technol.* 20, 407-413.
- Shi, Z. A., Baker, C. I., Lee, S. H., Park, S. A., Kim, S. C., & Ricke, S. U. (2016). Comparison of methods for quantitating *Salmonella enterica* Typhimurium and Heidelberg strain attachment to reusable plastic shipping container coupons and preliminary assessment of sanitizer efficacy. *J. Environ. Sci. Health, Part B.* 51(9), 602-608.
- US Food and Drug Administration. 2020. CFR 21 178.1010 Indirect Food Additives: Adjuvants, Production Aids, and Sanitizers, Subpart B – Substances Utilized to Control the Growth of Microorganisms.
- Wang, R., Zhou, Y., Kalchayanand, N., Harhay, D. M., & Wheeler, T. L. (2020). Effectiveness and Functional Mechanism of a Multicomponent Sanitizer against Biofilms Formed by *Escherichia coli* O157:H7 and Five *Salmonella* Serotypes Prevalent in the Meat Industry. *J. Food Protect.* 83(4), 568-575.

Wilson, C., Lukowicz, R., Merchant, S., Valquier-Flynn, H., Caballero, J., Sandoval, J., et al. (2017). Quantitative and qualitative assessment methods for biofilm growth: A mini-review. *Res. Rev. J. Eng. Technol.* 6(4), e-ISSN:2319-9873.

Yang, Y., Mikš-Krajnik, M., Zheng, Q., Lee, S., Lee, S., & Yuk, H. (2016). Biofilm formation of *Salmonella* Enteritidis under food-related environmental stress conditions and its subsequent resistance to chlorine treatment. *Food Microbiol.* 54, 98-105.

Zhao, X., Zhao, F., Wang, J., Zhong, N. (2017). Biofilm formation and control strategies of foodborne pathogens: Food safety perspectives. *RSC Adv.* 7, 36670-36683.

Table 1. Search keywords

Bacteria				Surface Type		Sanitizer
<i>Listeria monocytogenes</i> OR		Biofilm OR		Surface OR		Sanitizer OR
<i>Salmonella</i> (non-typhoidal) OR	AND	Attachment OR	AND	Coupon OR	AND	Hypochlorite OR
Shiga toxin-producing <i>E. coli</i>		Adhesion OR		Plastic OR		Chlorine dioxide OR
		Carrier		Stainless Steel OR		Quaternary ammonium compounds OR
				Rubber OR		Ethanol OR
				Wood OR		Iodophor OR
				Metal OR		Peroxyacetic acid
				Cement		

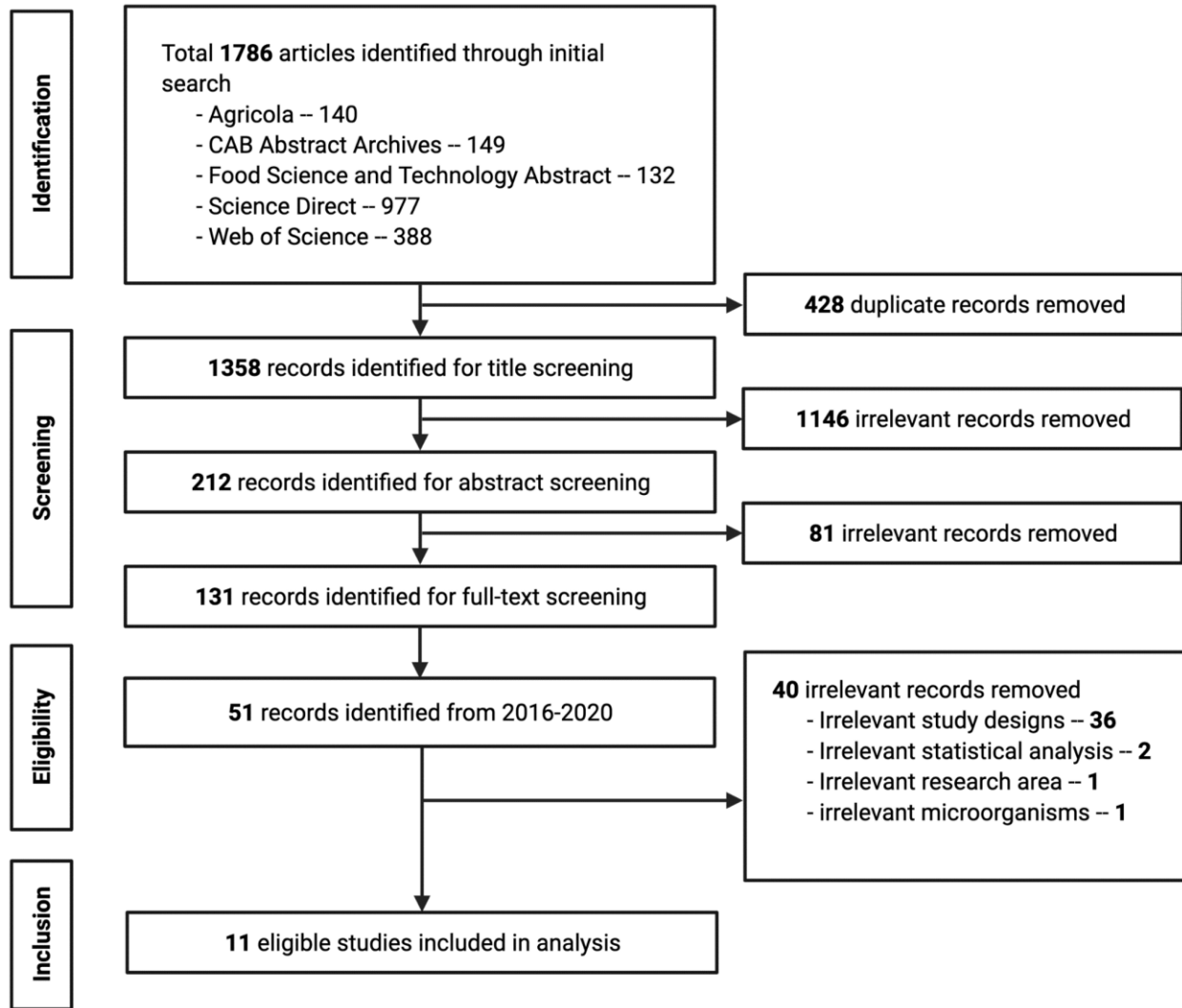


Figure 1. Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow chart describing the literature search procedure.

Table 2. Summary of the 11 peer-reviewed records from 2016-2020 included in the systematic literature review

Bacteria	Surface Type	Sanitizer Type	Sanitizer Concentration (ppm)	Sanitizer Contact Time (min)	Biofilm Community	Biofilm Formation Conditions	Organic Matter	Study Summary
<i>E. coli</i> O157:H7 ATCC 35150, 43889, 43890 (cocktail), <i>L. monocytogenes</i> ATCC 15315, 19114, 19115 (cocktail), <i>S. Typhimurium</i> ATCC 19585, 43971, DT104 (cocktail)	Stainless Steel	BZK* ¹ , H ₂ O ₂ , Iodophor, NaOCl,	20, 50, 100	5, 15, 30	SS*	24 hr, 25C, TSB	No	Majority of lar reductions are concentrations ppm with a co minutes or hig reductions ove from .12 to 2.2 log reduction v ppm NaOCl sa
<i>L. monocytogenes</i> ATCC 19113 (serotype 3b, human isolate)	Stainless Steel	NaOCl	50,100, 150, 200	1	SS	24 hr, 30C, TSB	No	The higher the the more log r log reductions .34 to 1.55. Th reduction was NaOCl sanitiz
<i>L. monocytogenes</i> 02 (truck wash drains dairy plant), <i>L. monocytogenes</i> 01 (clinical isolate), <i>S. Typhimurium</i> 101, clinical isolate	Stainless Steel, Rubber	Chlorine, Anionic acid, QAC** ² , Iodine	25, 100, 200	1, 2, 10	SS	48 hr, 22C, Tryptose phosphate broth, Peptone glucose	No	High concentr and Chlorine h amounts of log The log reduct from 0 to 5.6. reduction, 5.6, 100ppm Chlor

							phosphate broth		and the second reduction, 5.5, 200ppm QAC
<i>S. Typhimurium</i> ATCC 14028, <i>S. Enteritidis</i> ATCC 49216, <i>S. Typhimurium</i> ATCC 33062, <i>S. Senftenberg</i> 1734b	Polyurethane, Stainless Steel, Glass	NaOCl	40, 50, 60	10	SS		24 hr, 22C, TSB	Yes and No (Chicken Juice)	There was an present in some others. The ranged from 1 5.6 log reduction 40ppm NaOCl
<i>S. Heidelberg</i> SL486, <i>S. Heidelberg</i> SL486 marker (naldixic acid resistance), <i>S. Typhimurium</i> ATCC 14028	Plastic	PAA*** ³ , NaOCl	200	0.5	SS		24 hr, 37C, TSB	Yes (Bovine Serum Albumin)	200 ppm NaO more bacteria than PAA. The organic load p reductions ran 2.7. The 2.7 lo was from 200p sanitizer.
<i>S. Enteritidis</i> 124, <i>S. Enteritidis</i> 125, <i>S. Enteritidis</i> ATCC 13076	Stainless Steel	NaOCl	50	1	SS		48 or 168 hrs, 4 or 25 hrs, TSB	No	Highest log red with the biofil 48hrs in 4 C. T reductions ran 5.5. The 5.5 lo was from a 50 sanitizer.

<i>S. Typhimurium</i> FMCC B-137, FMCC B-193, FMCC B-415	Stainless Steel	NaOCl, PAA, BZK	10, 50	6	SS & MS** (<i>Staphylococcus aureus</i>)	144, 20C, TSB	No	The SS biofilm to remove for comparison to log reductions to 3.3. The largest reduction, 3.3, 50ppm BZK s
<i>S. Enteritidis</i> ATCC 13076, <i>S. Typhimurium</i> ATCC 14028	Stainless Steel	NaOCl, QAC	50, 200	1	SS & MS (<i>Pseudomonas aeruginosa</i>)	48, 96, 144 hr; 25C; TSB	No	The log reduction from 2 to 5.5. NaOCl had the reduction of 5. 200 ppm QAC largest log red
<i>L monocytogenes</i> SSA 151, serotype 1/2a	Stainless Steel	QAC	200	5	SS	24, 96, 168, 336 hr, 4, 15C; diluted TSB or Salmon broth	Yes and No (Salmon Broth)	The SS salmon largest log red biofilms were remove for the comparison to log reductions 1.2 to 7.4. The reduction was QAC sanitizer largest reduction studies.
<i>S. Enteritidis</i> phage type 8, <i>S. Enteritidis</i> phage type 8 pre-exposed 20 ppm QAC 120 h, 20C	Stainless Steel	QAC	200	5, 10, 15	SS & MS (<i>Pseudomonas fluorescens</i>)	48 hr, 20C, TSB	No	The 15 minutes reduced the biofilm was also the longest time. The SS biofilm easier to remove sanitizers in comparison to the MS. The log reductions ranged from 1

largest log red
from 200 ppm

<i>E. coli</i> O157:H7 strain 110, 141, 144, 168, 170, S. Dublin strain 519, S. Anatum strain 574, S. Montevideo strain 570, S. Newport strain 534, S. Typhimurium strain 554	Stainless Steel, PVC	QAC	300	10, 60, 360	SS	72 hr, 22- 25C, LB broth w/o salt	No	The 360 minute contact time re bacteria the m amount of sam time is too larg commitment f industry. The l ranged from 1 largest log red from 300ppm
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BZK*-Benzalkonium chloride; QAC**-Quaternary ammonium; PAA***-Peracetic acid