National Collaborating Centre
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Effectiveness of Alternative Antimicrobial Agents for Disinfection of Hard Surfaces

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evidence review

Summary

- A review of alternative antimicrobial agents reveals the need for standardized methodology for efficacy testing as well as considerations of toxicity, safety, cost, ease of use, availability, storage, and application-specific testing.

- The appropriateness of alternative antimicrobial agents, such as vinegar, lemon juice, and baking soda appear to be limited for commercial disinfection or sanitization, but some emerging technologies such as ozonated water and electrolyzed water have demonstrated substantial antimicrobial properties.

- Agents such as tea tree oil may demonstrate notable antimicrobial efficacy, but toxicity and lack of testing on hard surfaces limit their applications for hard surface disinfection. Thyme oil exhibits low toxicity and has been shown to be microbicidal, but its use may be limited due to the need for long contact time and costs.

- Although lacking active microbicidal activity, microfibre fabrics have unique properties that significantly increase their ability to remove organic debris (e.g., dust, bacteria, spores) and have the potential to be more efficient and economical than conventional cotton fabrics.

- Silver has been demonstrated to show residual antimicrobial properties. Its effectiveness in making materials/surfaces resistant to microbial growth has potential implications for expanding its use in medical and commercial applications.

- Further research is needed to explore potential uses of alternative agents in formulating novel disinfectants with desirable characteristics (e.g., lower toxicity, economical, environmentally friendly).

Introduction

Many alternative antimicrobial agents claim to exhibit comparable disinfection qualities to traditional disinfectants and sanitizers,[a] such as accelerated hydrogen peroxide, quaternary ammonium compounds (QUATs), and chlorine-based disinfectants (bleach). The alternative agents are often promoted as less toxic, environmentally friendly, and natural. The need for disinfectants as part of sanitation procedures has been supported by studies that show

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[a] For a discussion of traditional disinfectants and sanitizers, including definitions, please see the NCCEH evidence review on *Disinfectants and Sanitizers for Use on Food Contact Surfaces*.

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cross-contamination risks from environmental and food contact surfaces are not adequately reduced by the use of detergents and washing alone.[1]

This document is intended for public health inspectors and reviews the effectiveness, disinfection potential, and pertinent issues of major types of alternative agents that claim to have antimicrobial properties. Alternative agents that are reviewed include: tea tree oil, thyme oil, electrolyzed water, ozonated water, silver-based products, vinegar (acetic acid), lemon juice (citric acid), baking soda (sodium bicarbonate), and microfibre cloths. Table 1 summarizes the advantages and disadvantages of each alternative agent reviewed.

Unlike registered disinfectants, many alternative agents do not have a drug identification number (DIN).

The lack of a DIN indicates that product safety and effectiveness have not been formally reviewed and approved by Health Canada. Therefore, it may be difficult for public health inspectors (PHIs) to advise the public on the efficacy and safety of these alternative agents. Although uncommon, some alternative agents, such as thyme oil, silver, and citric acid are primary active ingredients in approved hard surface disinfectants. However, it is important to note that the antimicrobial efficacy of these alternative agents may be potentiated by other chemical compounds present in such registered disinfectants. Therefore, evaluating the efficacy of standalone alternative agents is likely not representative of results obtained using products in which a combination of ingredients, in addition to an alternative agent, is tested. Registered disinfectants can be found in Health Canada's _Drug Product Database_.[2]

Table 1. _Summary of notable advantages and disadvantages of alternative antimicrobial agents_[b]

Alternative agent	Advantages	Disadvantages	Primary active ingredient of at least one Health Canada registered disinfectant	Conclusions
Tea tree oil	• Natural product • Defined International Standards for composition of tea tree oil • TTO is used in existing topical medicinal treatments • No special equipment required to use	• Significant oral toxicity • May cause adverse skin reactions • Insoluble in water (may leave film of oil if used on hard surfaces)	• No	• Effective antimicrobial, but oral toxicity and hydrophobic properties limits its use as a sanitizer
Thyme oil[c]	• Natural product • Generally Recognized as Safe (GRAS status) • Low toxicity • Environmentally friendly	• Some bacteria are resistant to thyme oil (e.g., _P. aeruginosa_, _S. aureus_) • Expensive • Requires long contact-time (10 minutes)	• Yes	• Promising antimicrobial properties for use as a sanitizer • High cost may limit uses for large-scale applications
Electrolyzed water (EO water)	• Only salt and water required for production of EO water • On-site generation eliminates need for transport, storage, and	• Acidic EO water has corrosive properties • Safeguards are required as chlorine gas produced in production	• No	• Promising antimicrobial properties for use as a sanitizer • Potential to be used for large-scale

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[b] A brief discussion, including references, for the advantages and disadvantages in this table is available within the reviews for each alternative antimicrobial agent.
[c] Thymol is no longer listed as an asthmagen by the Association of Occupational and Environmental Clinics (AOEC)[3]

Alternative agent	Advantages	Disadvantages	Primary active ingredient of at least one Health Canada registered disinfectant	Conclusions
	handling of hazardous chemicals • Abundantly and readily produced • Low operating costs • No toxic/chemical residues left on surfaces	chambers • High startup and maintenance costs (special equipment for production and dispensing required) • Rapid dissipation of antimicrobial activity		applications
Ozonated water (aqueous ozone)	• Only oxygen (e.g., in air or compressed) required for production • On-site generation eliminates need for transport, storage, and handling of hazardous chemicals • Devices have been registered with NSF International and the Canadian Food Inspection Agency • U.S. Food and Drug Administration has approved ozone (gas and aqueous phase) as an antimicrobial • Maintains efficacy in cold water • Abundantly and readily produced • No toxic/chemical residues left on surfaces	• High startup, operating, and maintenance costs (special equipment for UV or corona discharge, dispensing, etc.) • Potential occupational exposure to ozone • Damaging to sensitive materials • Rapid dissipation of antimicrobial activity	• No	• Promising antimicrobial properties for use as a sanitizer • Potential to be used for large-scale applications
Silver	• Existing uses of silver in drinking water, swimming pools, medical devices • Numerous potential applications for silver-impregnated materials/ nanotechnology • Demonstrated residual antimicrobial activity	• Slow-acting antimicrobial • Microbial resistance has been identified • Interference by proteins and salts • Low toxicity at levels needed for antimicrobial activity • Loses antimicrobial properties once all silver ions have been released	• Yes	• Research shows potential for numerous applications as an antimicrobial agent • More research is needed to define the parameters required to be effective • Applications may be limited to residual antimicrobial activity (i.e., non-immediate uses)

Alternative agent	Advantages	Disadvantages	Primary active ingredient of at least one Health Canada registered disinfectant	Conclusions
Vinegar (acetic acid) Lemon juice (citric acid) Baking soda (sodium bicarbonate)	• Natural product • Readily available and abundant • Low toxicity	• Limited antimicrobial efficacy and narrow in spectrum • May damage the organoleptic properties of produce • May be corrosive or irritating • Has pungent and unwanted odours • Mixing acids with bleach can cause the production of chlorine gas	• Acetic acid: No • Citric acid: Yes • Sodium bicarbonate: No	• Applications are limited by poor antimicrobial efficacy and aesthetic considerations • Potential to be used in formulations of disinfectants • Unlikely to be used for commercial applications, but may have uses in domestic settings
Microfibre	• Readily available • More effective at cleaning than cotton fabrics • Lighter material – can promote productivity and reduce occupational injury • May minimize the use of chemicals • Can be cost effective	• Lacks active antimicrobial properties – may become a source of contamination for subsequently cleaned surfaces • Damaged by heat, chlorine-based disinfectants, and fabric softeners • More expensive than cotton	• No	• Promising efficacy for cleaning, but not as an antimicrobial

Tea Tree Oil

This essential oil, extracted from the leaves of *Melaleuca alternifolia*, is widely used as an alternative antimicrobial agent and international standards for the composition of tea tree oil (TTO) have been developed (e.g., ISO 4730).[4] It is often used as a topical anti-inflammatory agent and to treat skin infections such as acne, ringworm, scabies, and athlete's foot.[5,6]

The hydrophobic properties of TTO are hypothesized to impair cell membrane integrity. Supporting studies have revealed the effects of TTO on bacterial and fungal cells, demonstrating the leakage of intracellular components, inhibition of cellular respiration, and an increase in susceptibility to sodium chloride.[5,7,8] Available research has suggested the potential for antiviral and antiprotozoal activity, but studies have been limited in scope.[5] Terpinen-4-ol has been noted

as the primary antimicrobial agent in TTO, but several other components are also microbicidal or facilitate antimicrobial activity.[5,7]

Antimicrobial efficacy

Researchers have used European Standards for evaluating the use of TTO as a sanitizer for food areas (EN 1276) and as an antiseptic agent for hand washing (EN 12054).[9] The minimum standard is a 5 log reduction in 5 minutes for use as a sanitizer and a 2.52 log reduction in 1 minute for use as a hand washing agent. Test suspensions of *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa* were treated with 1% to 10% (v/v) TTO and log reductions were recorded after 1 minute and 5 minutes of

treatment.[9] Treatment with 5% TTO resulted in a 5 log reduction of *E. coli* in 1 minute and a 4 log reduction of *P. aeruginosa* in 5 minutes. Treatment with 8% TTO resulted in a 5 log reduction of *P. aeruginosa* in 1 minute. Log reductions of *S. aureus* ranged from 0.19 (1% TTO, 1 minute) to 0.80 (10% TTO, 1 min) and did not significantly differ with varying concentrations of TTO or contact time.[9] As an antiseptic hand wash agent, 2.75% TTO resulted in the reduction of *E. coli* and *P. aeruginosa* by 4 logs and 2 logs, respectively, in 1 minute; the same treatment resulted in a log reduction of <0.5 for *S. aureus*.[9] Other studies have determined minimum inhibitory concentrations for *E. coli* and *S. aureus* to be 0.25% and 0.50% (v/v), respectively.[10]

Potential use for disinfection: applicability and pertinent issues

Ingestion of undiluted TTO may induce temporary neurological effects in children and adults. Symptoms include confusion, inability to walk, disorientation, ataxia, unconsciousness, and coma.[6,11] Allergic skin reactions, systemic reactions, and irritation are also associated with dermal exposure to TTO. The LD_{50} of tea tree oil is estimated to be 1.9-2.6 mL/kg in rats and when used undiluted has been determined to be unsafe by scientific committees of the European Commission; it can be irritating to the skin at concentrations as low as 5% (v/v).[12] Therefore, uses of TTO on food contact surfaces and in settings with sensitive individuals may be limited by its oral and dermal toxicity to humans.

There also exists preliminary evidence that indicate the potential for TTO as an endocrine disruptor, altering the signalling of sex hormones that affect human development.[13] In particular, one clinical report has suggested that pre-pubertal gynecomastia, the abnormal growth of breast tissue in preadolescent males, may be related to the repeated use of personal products containing lavender oil and/or lavender oil combined with TTO (both are essential oils).[14,15] Investigations through mammalian cell culture studies have revealed that TTO has slight "estrogenic and antiandrogenic activities".[13,14,16] However, another study emphasizes the need to consider bioavailability when conducting human health risk assessments and that the aforementioned *in vitro* results are unlikely to represent *in vivo* human health risks.[16] Results from this study supported the *in vitro* estrogenic and antiandrogenic effects of TTO, but also illustrated that TTO components, known to penetrate the skin, do not induce measurable effects.[16] From this, the report

suggests that further human health risk assessments regarding TTO should characterize TTO components and their bioavailability, in conjunction with observations for possible estrogenic and/or antiandrogenic properties. Furthermore, threshold levels of TTO required to induce these effects have yet to be characterized. More evidence is required in order to evaluate these potential health effects and their significance, if any, to the population and to public health.

Thyme Oil

Thymol and carvacrol are the main components in thyme oil that are believed to exhibit the most antimicrobial activity.[17] They have been demonstrated to cause an increase in permeability of the cell membranes of bacteria, a reduction in the proton motive force, and an associated decrease in intracellular levels of adenosine triphosphate (ATP, a high-energy molecule responsible for providing energy to drive reactions in the cell).[18,19] Although there is insufficient evidence to support its effectiveness for health benefits, thyme oil has been taken orally to treat sore throat, cough, bronchitis and inflammatory conditions of the gastrointestinal tract.[20] As a topical agent, it can be used as an anti-inflammatory mouthwash and for treatment of ear infections. Thyme oil is also a food additive and, in the United States, is Generally Recognized as Safe (GRAS) for ingestion (21 *CFR* 182.10).[21]

Antimicrobial efficacy

At concentrations of 0.1 to 0.6% (v/v), thyme oil is shown to inhibit the growth against microorganisms, such as *Campylobacter jejuni, E. coli* O157:H7, *Listeria monocytogenes, Salmonella spp.*, *S. aureus*, and *Candida albicans*; however, much higher concentrations (e.g., 2 to 10%) are needed to be bacteriostatic against *P. aeruginosa*.[17,22-24] When compared to tea tree oil, thyme oil has been shown to have a lower minimal inhibitory concentration (MIC) to a variety of microorganisms.[23,24] A 5 log reduction in *E. coli* was observed within 5 minutes of exposure to 0.31% thyme oil whereas the same reduction in *S. aureus* took 15 minutes with 2.5% thyme oil.[24] Populations of *P. aeruginosa* did not achieve this reduction even after 24 hours of exposure to >10% thyme oil.

Potential use for disinfection: applicability and pertinent issues

Thyme oil is natural, environmentally friendly, and has been used as a primary active ingredient in several disinfectant products registered with Health Canada. Classified as a *Minimum Risk Pesticide*, it has low oral and dermal toxicity, allowing it to be exempt from some sections of the U.S. *Federal Insecticide, Fungicide, and Rodenticide Act* and pesticide registration requirements.[25] Also, some thymol-based registered disinfectant products do not require a rinsing or wiping step for disinfecting surfaces and can be safely used undiluted.[26] Furthermore, the long contact times for required disinfection (e.g., 10 minutes) may inhibit its use for large-scale applications.

Electrolyzed Water (Electrolyzed Oxidizing Water)

Although the mechanism has not been fully described, this method of disinfection has been hypothesized to rely on the chlorine-based disinfection properties of hypochlorous acid (free chlorine) produced by the electrolysis of a salt (sodium chloride) solution.[27] In addition, studies have shown that electrolyzed oxidizing water (EO water) is more effective at inactivating microbes than chlorine solutions with similar free chlorine concentrations, suggesting that oxidation reduction potential (ORP) and low pH, in addition to free chlorine, may be synergistic to the antimicrobial activity of EO water.[28] Typically, acidic EO water has a free chlorine level of 10 to 90 ppm, ORP of 1100 mV, and a pH of 2 to 3.[27,29] Neutral (pH 6 to 8) and alkaline (pH 10 to 13) forms of EO water can also be produced by increasing the concentration of hypochlorite ions (OCl^-); this may be done to reduce its corrosiveness.[27]

Antimicrobial efficacy

EO water has been tested for efficacy of use in numerous applications, such as washing produce, decontamination of egg shells (>6 log reduction in *Salmonella enteritidis* in 1 minute), and decontamination of hides of cattle (3.5 log reduction in aerobic plate count, 4.3 log reduction in *Enterobacteriaceae* count, 47% reduction in number of hides testing positive for *E. coli* O157:H7).[30-32] EO water has been shown to be effective at inactivating a variety of microorganisms of public health significance. Suspensions of *E. coli* O157:H7, *S. enteritidis*, *P.*

aeruginosa, *C. jejuni*, *S. aureus*, *L. monocytogenes* have shown to be inactivated by approximately 7 logs in 1 minute or less after treatment with EO water.[28,33,34] The efficacy of EO water on food contact surfaces, produce, poultry, fish, and pork have also been reviewed.[29] Summary of the findings indicate test organisms were reduced by log reductions of 2.0-6.0 for hard surfaces/utensils, 1.0-3.5 for vegetables/fruits, 0.8-3.0 for chicken carcasses, 1.0-1.8 for pork, and 0.4-2.8 for fish.[29] These reductions represented treatments with contact times ranging from less than 1 minute to 20 minutes, in some cases.[29] Furthermore, washings obtained from inoculated stainless steel and glass surfaces, after treatment with EO water, have been found to contain <1 log CFU/mL of test organisms, illustrating the potential for EO water to reduce cross contamination from the processing water.[35]

Potential use for disinfection: applicability and pertinent issues

Besides the antimicrobial efficacy of EO water, the low operating costs from the availability of salt and water make EO water a promising alternative disinfectant. However, there is a high initial cost for installing special equipment to produce and dispense EO water. As no residue or noxious gas remains after application of EO water, the agent is noted to be environmentally and worker friendly.[32] Also, on-site generation of EO water eliminates the need for special transportation, handling, or storage of hazardous chemicals.[29] However, rapid dissipation of antimicrobial activity may prevent EO water solutions from being stored in an open environment for extended periods. Corrosion of certain metals (e.g., carbon steel, copper) has also been noted in certain studies; this can be minimized by using neutral EO water.[36] Furthermore, chlorine gas may be generated in the anode chamber during production of EO water and the appropriate safeguards and response measures must be present should leakage occur.[27] The toxicity of EO water has also been noted as lower than that of conventional disinfectants and no adverse oral or digestive health effects were observed in mice given EO water as drinking water.[32,37]

Similar to chlorine-based sanitizing treatments, microbial reduction may vary depending on the susceptibility of test organisms, contact time, treatment methodology, presence of organic residues/debris, and the surface/texture of the area being sanitized. The presence of organic residues, uneven textures, and porous surfaces has been demonstrated to impair the antimicrobial efficacy of EO water.[38,39] Studies have

also shown inactivation of microbes to be dependent on temperature of EO water, for example, EO water at 45°C favours microbial inactivation when compared to EO water at 23°C.[33]

Ozonated Water (Aqueous Ozone)

Ozone (O_3), a gaseous oxidizing agent with antimicrobial properties, can be generated on-site and dissolved into water to create an ozone enriched water solution. Potential uses of ozonated water include washing and extending the shelf life of produce, decontaminating and lowering the chemical oxygen demand of processing waters, sanitation of hard surfaces, and decontamination of cattle hides.[31,40-43] However, ozone is highly unstable and has limited solubility in water.[42] Therefore, the antimicrobial activity of ozonated water dissipates rapidly and may limit its applications for non-immediate uses.

In 2001, the U.S. Food and Drug Administration approved ozone to be used as an "antimicrobial agent" and in the "treatment, storage, and processing of foods" as outlined in the U.S. *Code of Federal Regulations* (21 *CFR* 173.368).[44] NSF international has registered devices deemed "acceptable for use as an ozone generating device providing sanitation and disinfection to hard, inanimate, pre-cleaned surfaces, in and around food processing areas."[45] Furthermore, the Canadian Food Inspection Agency (CFIA) has deemed several devices that produce ozonated water for sanitizing hard surfaces as acceptable for use in establishments under their regulatory authority (i.e., a federally registered food processor). Such devices are listed in the searchable database, *Reference Listing of Accepted Construction Materials, Packaging Materials and Non-Food Chemical Products*.[46]

Antimicrobial efficacy

Lettuce dipped in ozonated water (4 ppm O_3, 20°C) for 2 minutes had significant reductions in *Enterobacteriaceae* (1.3 log CFU/g) as well as psychrotrophic (1.5 log CFU/g) and mesophilic bacteria (1.7 log CFU/g).[43] These results were comparable to treatment with 100 ppm chlorine in the same conditions and researchers have suggested that ozonated water may be a potential alternative to chlorine dippings.[43] Use of ozonated water in place of chlorine in produce processing may avoid the formation of undesirable chlorine disinfection by-products (e.g.,

trihalomethanes). When used to decontaminate cattle hides, ozonated water was able to achieve a 2.1 log reduction in aerobic plate count, 3.4 log reduction in *Enterobacteriaceae* count, and 58% reduction in number of hides testing positive for *E. coli* O157:H7.[31]

Researchers have also used European Standards EN 1040 and EN 1275 to determine the bactericidal and fungicidal efficacy of ozonated water.[47] Test suspensions of *S. aureus*, *E. coli*, *P. aeruginosa,* and *Enterococcus hirae* were inactivated (>5 log reduction) in 30 seconds when treated with ozonated water with a concentration of 3 ppm O_3; test suspensions of *C. albicans* were inactivated (>4 log reduction) under similar treatment conditions.[47] No reduction in the number of viable spores of *Aspergillus brasiliensis* was observed, even after treatment with ozonated water (1.5 to 3 ppm O_3) for 30 minutes. Furthermore, a reduction in approximately half of the ozone concentration (e.g., 3.0 ppm to 1.5 ppm) was observed after 30 minutes of storage. Lower concentrations of ozone (e.g., 0.15 to 0.20 ppm O_3) in water can achieve comparable log reductions but contact time of 1-5 minutes is required.[48]

Potential use for disinfection: applicability and pertinent issues

Ozonated water has strong non-selective antimicrobial properties, leaves no chemical residues, can be used with cold water, and can be produced on demand. Furthermore, on-site generation of ozonated water avoids the need for special transportation, handling, and storage of hazardous chemicals. However, in order to produce ozonated water, high energy UV radiation (e.g., 188 nm wavelength) or electrical discharges (e.g., corona discharge) are required to convert oxygen in the air (or pure oxygen) into ozone gas.[42] These processes require special equipment and substantial amounts of electrical energy to operate, leading to high initial and operating costs for large-scale commercial applications.

Limited information exists on the toxicity of ozonated water, but studies have shown no significant adverse effects on human oral epithelial cells after acute exposure.[49] Other potential limitations for its use include damage to sensitive materials (e.g., rubber gaskets) and occupational safety associated with exposure to ozone gas.[42,49]

Silver

Studies have shown that the likely modes of microbial inactivation by silver is through interference with cellular respiration and transport, interactions with DNA, disruption of proteins, and destruction of the cell membrane.[50] Contrary to many disinfectants, potential applications of silver are commonly associated with slow release of silver from silver-impregnated materials and residual antimicrobial effects.[51]

Silver has been used for its antimicrobial properties in drinking water/cooling tower disinfection, swimming pools, and for medical uses.[52] Notably, Health Canada has issued a DIN for a silver dihydrogen citrate-based disinfectant (silver dihydrogen citrate 0.003% and citric acid 4.846%) for use as a hard surface disinfectant with demonstrated residual activity.[53]

Antimicrobial efficacy

Antimicrobial efficacy of silver-impregnated packaging liners on spoilage organisms from meats and melons has also been evaluated.[54,55] For meat liners, an average difference of 1 log CFU/g was observed between silver-impregnated pads and control pads. For melon liners, an average difference of 3 log CFU/g was observed between silver-impregnated pads and control pads. As silver has an affinity for proteins and salts, meat exudates likely interfere with the antimicrobial activity of silver to a greater extent than melon juices.[55]

Several silver-impregnated wound dressings have also been evaluated for bactericidal efficacy.[56] Notably, antimicrobial activity may depend on release rate of silver from the impregnated material, as well as the matrix type. For example, it has been demonstrated that a 24-hour silver release rate of approximately 93 ppm can result in >3.46 log reduction (30 min contact time) in *S. aureus*.[56] However, with a dressing of a different matrix type, no log reduction was observed (30 min contact time) even though the dressing had a higher 24-hour silver release rate of 318 ppm.

In one study, stainless steel coupons and cups were coated with a silver-zinc zeolite (2.5% w/w silver and 14% zinc), then inoculated with test organisms (e.g., *S. aureus*, *E. coli*, *P. aeruginosa*, and *L. monocytogenes*) to evaluate bactericidal activity by recovery of organisms at different time intervals (e.g., 0h, 4h, 24h). Microbial reduction of up to 5 logs was observed in 24 hours, when compared to untreated controls.[57] Microbial reductions observed at 4h diminished after 5 washings with a towel, but reductions at 24h remained >90% after 11 washings.[57] A similar study using the same silver-zinc zeolite coatings showed that the numbers of vegetative cells of *B. cereus* were reduced by 3 logs after 24h, but spores were not inactivated even after 48 hours.[58]

Furthermore, an alcohol-based (79%) disinfectant spray with silver iodide (0.005%) was tested for residual bactericidal activity. When compared to untreated controls, populations of *P. aeruginosa* and *S. aureus* were reduced by >3 logs in 2 hours and by >4 logs in 8 hours; a chlorine based disinfectant showed similar residual activity, but only with *S. aureus*.[59] Multiple rinses, abrasion, and re-contamination did not affect residual activity.

Potential use for disinfection: applicability and pertinent issues

Accumulation of silver in the body may lead to side effects including: impaired absorption of medicine, neurological problems, kidney damage, headache, fatigue, and skin irritation.[60] However, the levels of silver in silver-based antimicrobial products have not been documented to cause adverse effects in humans and are unlikely to cause the side effects associated with chronic ingestion of high levels of colloid silver products, which may lead to argyria (an irreversible condition which manifests as blue discolouration of the skin and/or eyes).[60-62] Silver has no known function in the body and health claims associated with the use of colloid silver products have yet to be substantiated.[60] For a discussion on nanosilver technologies, please see the NCCEH contracted review by Green and Ndegwa (2011) titled: *Nanotechnology: A Review of Exposure, Health Risks, and Recent Regulatory Developments*.[63]

The World Health Organization (WHO) has suggested that the NOAEL of silver in drinking water to be 10 g consumed over a lifetime (i.e., daily ingestion of 2 L of water containing 0.2 mg/L silver for 70 years).[64] Importantly, the levels of silver in natural waters and drinking water have been noted to be thousands, if not millions, of times lower than the NOAEL (e.g., 5 µg/L).[64]

Other issues noted with the use of silver as an antimicrobial include: the emergence of silver-resistant microbes (e.g., cellular efflux pumps), interference from proteins and salts, current lack of standardization for efficacy testing, and loss of antimicrobial properties once all active silver is released from impregnated

materials.[52,65-67] Also, the slow-acting antimicrobial activity of silver limits its use for immediate disinfection of hard surfaces. Nevertheless, the residual antimicrobial activity of silver may allow for potential applications in formulations of disinfectants or to reduce the harbourage of microbes on surfaces and subsequent transfer of microbes from one surface to another.

Vinegar (acetic acid), Lemon Juice (citric acid), and Baking Soda (sodium bicarbonate)

The antimicrobial properties of vinegar and lemon juice are commonly associated with their acetic acid and citric acid content, respectively.[68] These organic acids are hypothesized to cross the cell membrane of bacteria where the release in protons (H^+) causes the cells to die.[69] As the growth of many pathogenic organisms are inhibited in conditions where the pH is <4.6, these organic acids, with a pH 2 to 3, are commonly added to foods as a preservative.[69]

Baking soda has been used to formulate toothpaste, cosmetic products, and is known for its acid-neutralizing properties, but limited peer-reviewed evidence exists for its antimicrobial activity on hard surfaces.[70] It has been reported to be virucidal and inhibit the growth of several fungi, but its mechanism of action is unclear.[71,72] Baking soda has also been shown to enhance the effectiveness of other agents for controlling mould growth on produce, but its antifungal spectrum may be limited.[73-77] In addition, because the pH of baking soda in a neutral solution equilibrates at a maximum near 8.34, its pH alone is likely insufficient to inhibit the growth of many foodborne microorganisms, many of which can grow in conditions with up to pH 9 to 10.[70,78] However, at least one study has suggested its chemical properties (e.g., alkaline pH, mild abrasive) can be effective for cleaning kitchen surfaces.[79]

Antimicrobial efficacy

Numerous studies have demonstrated the antimicrobial efficacy of acetic acid (AA), citric acid (CA), and sodium bicarbonate (SB) using suspensions of bacteria, recovery from treated hard surfaces, rinsing meat, and washing produce (summarized in Appendix A).[68,80-92] However, it is difficult to compare results between studies as there are no standardized experimental parameters used to test efficacy. Notably, efficacy demonstrated in suspensions are drastically different from efficacy demonstrated on produce (i.e., in the absence or presence of organic matter).

Results from studies suggest that vinegar (acetic acid) exhibits the most antimicrobial efficacy, followed by lemon juice (citric acid) and baking soda (sodium bicarbonate).[68,90] Typically, Gram-negative bacteria, such as *Shigella sonnei*, *Salmonella spp.*, *E. coli*, *P. aeruginosa*, and *Yersinia enterocolitia* are more susceptible to organic acids (e.g., acetic acid, citric acid) than Gram-positive bacteria, such as *S. aureus* and *L. monocytogenes*. The highly cross-linked cell walls of Gram-positive bacteria are believed to impair the diffusion of the organic acids into the cell, preventing antimicrobial action.[68,69,83] Baking soda is generally ineffective against *E. coli, P. aeruginosa, S. aureus*, and *Salmonella spp.*, but had notable virucidal activity against feline calicivirus (norovirus surrogate).[68,72,89] The efficacy of AA, CA, and SB vary greatly (<1 log to >5 log reduction in test microbes; contact times ranged from 0.5 min to 15 min) depending on test organisms and test conditions. When used at higher temperatures, vinegar and lemon juice are observed to result in increased antimicrobial efficacy.[68,91,93] The difficulty in assessing antimicrobial efficacy and narrow antimicrobial spectrum of these alternative agents may limit their applications as hard surface disinfectants.

Potential use for disinfection: applicability and pertinent issues

Vinegar, lemon juice, and baking soda have the advantage of being readily available, environmentally friendly, low in toxicity, and natural. Although these agents are commonly used to eliminate odours, residual odour and taste on surfaces may be unwanted in applications where public or occupational exposure is undesirable, especially at concentrations that exhibit antimicrobial activity. Organoleptic properties of produce washed with AA or CA may also be adversely affected (e.g., wilting or souring).[90] Furthermore, like chlorine-based disinfectants, the efficacy of organic acids (AA and CA) is drastically reduced in the presence of organic matter. Potential safety concerns are also noted with the use of vinegar, lemon juice, and/or baking soda for sanitation purposes. For example, if chlorine-based disinfectants (e.g., bleach) are used simultaneously with vinegar and/or lemon juice to sanitize hard surfaces, there is potential for an increased risk of accidental mixing, which may result in the formation of chlorine gas. Designated containers (e.g., spray bottles or buckets) with proper labelling

would be necessary, as with all chemical disinfectants, to indicate the contents and reduce the chance of mixing incompatible chemicals. The low pH of AA and CA can also make these agents a potential eye, nose, and respiratory tract irritant.

Overall, it is unlikely that vinegar, lemon juice or baking soda, by themselves, will become mainstream antimicrobial agents in commercial settings, but the notable efficacy of vinegar and lemon juice may have indications for their potential use as household antimicrobial agents or in formulations of disinfectants.

Microfibre Cloths

Microfibres are extremely fine strands of fibres that are less than 1 denier (i.e., a single strand weighs ≤ 1 gram per 9,000 metres). Due to the unique structure of the fibres, micrometre diameters, and electrostatic properties, the fabrics made from microfibres have an ability to trap dust and microbes more effectively than conventional cotton cloths or mops; this is likely attributed to the high surface area and capillary effect of microfibre fabrics.[94,95] Depending on the weave and composition of the fibres, properties of water absorption, permeability, stain-resistance, and wrinkle-resistance can vary.

Antimicrobial efficacy

The fibres themselves have not been shown to be microbicidal, but have been shown to demonstrate considerable cleaning efficacy, by physical removal of microbes and organic debris from surfaces.[95-97] For example, microfibre cloths (with water) have been documented to reduce *S. aureus*, *E. coli*, and *Clostridium difficile* spores on hard surfaces, by an average of 1 to 3 logs.[97,98] However, it is difficult to make general statements regarding efficacy due to the lack of standardized testing methods and manufacturing parameters for microfibre cloths. Even so, the application of microfiber cloths/fabrics for cleaning may help maximize the effectiveness of conventional antimicrobial products.

Potential use for cleaning: applicability and pertinent issues

Although microfibre cloths are more expensive than cotton cloths, the U.S. Environmental Protection Agency has a case study to demonstrate that the use of microfibre mops, in place of conventional mops in

hospital cleaning programs, can be more economical by saving on labour, chemical, water, and electrical costs.[99] However, the cleaning efficacy of microfibre is reduced through damage that can be caused by high heat (e.g., process temperatures of industrial washing machines), some disinfectants (e.g., bleach), and fabric softeners.[95,98,99] In addition, studies have emphasized that the lack of antimicrobial properties allows for the potential for cross-contamination or re-contamination of subsequently cleaned surfaces if used with water alone (i.e., transmission of diseases in institutional and food processing settings).[88,96,100,101]

Evidence Gaps

The emergence of new alternative antimicrobial agents and their use for disinfection requires further research and review. The current lack of standardized evaluation criteria makes it difficult to compare antimicrobial properties across different types of alternative agents. In particular, more research is required to better define the concentration, contact time, and stability required for these agents to induce antimicrobial effects, if any. In addition, defining the composition of alternative agents will help with comparison. If alternative agents are considered for use on food contact surfaces, the need for a final rinsing step must also be evaluated.

Further understanding of the antimicrobial mechanisms of alternative agents may help to define relevant properties for use as disinfectants. For example, how are they affected by organic residues or other chemicals? How can they be made or used more effectively? Do they possess potential or synergistic properties when combined with other disinfectants? Do they exhibit unique properties that are desirable (e.g., residual antimicrobial effects)? Further research to explore their potential uses in formulating novel disinfectants may help evaluate their role, if any, in manufacturing disinfectant products with desirable characteristics (e.g., lower toxicity, economical, environmentally friendly).

Acknowledgments

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Appendix A: Antimicrobial efficacy data of vinegar (acetic acid), lemon juice (citric acid), and baking soda (sodium bicarbonate)

Organism	Test conditions	Concentration (v/v, unless otherwise indicated), contact time (at 4 to 25°C unless otherwise indicated)	Log reduction (CFU/mL or g)	Ref.
Aerobic plate count	Microflora on lettuce	Vinegar (1.9% acetic acid), 10 min w/ agitation	2.3	(Vijayakumar & Wolf-Hall, 2002)[90]
		Lemon juice (0.6% citric acid), 10 min w/ agitation	1.8	
	Microflora on parsley	2 and 5% acetic acid solutions, 15 min	5	(Karapinar & Gonul, 1992)[92]
	Microflora on cilantro	Citric acid (0.6% prepared solution), 1 min	<1	(Allende, 2009)[81]
	Raw skinless/ boneless chicken breast	Vinegar (5% acetic acid), 1 min w/ agitation	2.2	(McKee et al., 2005)[83]
		Baking soda (10% sodium bicarbonate solution), 1 min w/ agitation	1.0	
Escherichia coli O157:H7	Suspension	Vinegar (5% acetic acid), 5 min	2.4	(Rutala et al., 2000)[89]
		Baking Soda (8% sodium bicarbonate), 5 min	0.7	
		Vinegar (5% acetic acid), 1 min at 55°C	>5.0	(Yang et al., 2009)[68]
		Citric acid (5% prepared solution), 10 min at 55°C	>5.0	
		Baking Soda (11, 33, and 50% sodium bicarbonate solution)	<1	
	Inoculated lettuce	Vinegar (5% acetic acid), 5 min	3.0	(Chang & Fang, 2007)[87]
	Inoculated cilantro	Citric acid (0.6% prepared solution), 1 min	<1	(Allende, 2009)[81]
Escherichia coli CDC1932 (nalidixic acid resistant strain)	Inoculated lettuce	Vinegar (1.9% acetic acid), 10 min	5.4	(Vijayakumar & Wolf-Hall, 2002)[90]
		Lemon juice (0.6% citric acid), 10 min w/ agitation	2.1	
Staphylococcus aureus	Suspension	Vinegar (5% acetic acid), 5 min	0.3-2.3	(Rutala et al., 2000)[89]
		Baking Soda (8% sodium bicarbonate), 5 min	0.5	
Salmonella choleraesuis	Suspension	Vinegar (5% acetic acid), 0.5 min	>6.0	(Rutala et al., 2000)[89]
		Baking Soda (8% sodium bicarbonate), 5 min	2.3	
Salmonella Typhimurium	Suspension	Vinegar (5% acetic acid), 1 min	>5.0	(Yang et al., 2009)[68]
		Citric acid (5% prepared solution), 1 min at 55°C	>5.0	
	Inoculated spring onion and rocket leaves	Lemon juice (4.2% citric acid), 15 min	2.95 (rocket leaves), 1.70 (spring onion)	(Yucel Sengun & Karapinar, 2005)[85]
		Vinegar (3.95% acetic acid), 15 min	2.20 (rocket leaves), 1.19 (spring onion)	(Yucel Sengun & Karapinar, 2005)[85]
	Inoculated carrots	Vinegar (4.03% acetic acid), 15 min	1.87	(Yucel Sengun & Karapinar, 2004)[84]
		Lemon juice (4.46 citric acid), 15 min	2.68	
	Inoculated stuffed mussels	Lemon juice (5.88% citric acid), 15 min	0.56	(Kişla, 2007)[82]

Organism	Test conditions	Concentration (v/v, unless otherwise indicated), contact time (at 4 to 25°C unless otherwise indicated)	Log reduction (CFU/mL or g)	Ref.
Pseudomonas aeruginosa	Suspension	Vinegar (5% acetic acid), 0.5 min	>5.8	(Rutala et al., 2000)[89]
		Baking Soda (8% sodium bicarbonate), 5 min	1.1	
Listeria monocytogenes	Suspension	Vinegar (5% acetic acid), 1 min at 55°C	>5.0	(Yang et al., 2009)[68]
		Vinegar (5% acetic acid), 10 min at 25°C		
		Citric acid (5% prepared solution), 10 min at 55°C		
Yersinia enterocolitica	Suspension	Citric acid (5% prepared solution), 10 min at 4°C	<1	(Virto et al., 2005)[93]
		Citric acid (5% prepared solution), 10 min at 20°C	<1	
		Citric acid (5% prepared solution), 2 min at 40°C	>4	
		Citric acid (10% prepared solution), 10 min at 4°C	<1	
		Citric acid (10% prepared solution), 10 min at 20°C	>4	
		Citric acid (10% prepared solution), 1 min at 40°C	>4	
	Inoculated parsley	Vinegar (1.96 and 2.45% acetic acid), 15 min	5	(Karapinar & Gonul, 1992)[92]
Shigella sonnei	Inoculated parsley	Vinegar (5.2% acetic acid), 5 min w/ agitation	>6.0	(Wu et al., 2000)[91]
Poliovirus	Suspensions	Vinegar (5% acetic acid), 5 min	0.32	(Rutala et al., 2000)[89]
		Baking soda (8% sodium bicarbonate), 5 min	0.42	
Feline calicivirus (norovirus surrogate)	Inoculated stainless steel disks	Baking soda (5% sodium bicarbonate), 1 min	4	(Malik & Goyal, 2006)

Appendix B. Search Methodology

Literature searches were conducted to locate articles that support a brief review and discussion of the mechanism of action, disinfection potential, pertinent issues, and safety/toxicity concerns of each alternative antimicrobial agent. Bibliographies of retrieved articles were scanned to further retrieve more extensive and detailed information on a particular subject of interest. Any related articles and suggested articles, appearing within the search engine, were also considered for inclusion. This process subsequently aided in refining search terminology and finding additional and specific articles of interest.

Inclusion of articles, with publishing dates from years 2001-2011, were preferable; articles were not excluded by date if their material was of particular interest or the date of publication did not adversely impact the quality of evidence. Grey literature was included for descriptive and illustrative purposes.

Search engines/databases for sources of information

- University of British Columbia Library – 'Summon' Search (publisher list here)
- Pubmed
- ScienceDirect
- Ingentaconnect
- MedlinePlus.

Search terminology

Names of each antimicrobial agent, including any alternative names or parts of names, were used by themselves or in combination with:

- Action
- Activity
- Advantage
- Adverse
- Antimicrobial
- Applica*
- Bacteri*
- Biocid*
- Characteristic
- Potential
- Propert*
- Review
- Disadvantage
- Disinfect*
- Effectiv*
- Efficac*
- Food
- Germicid*
- Health effect
- Mechanism
- Microbicid*
- Safety
- Surface
- Toxic*

Each alternative agent was reviewed on the basis of antimicrobial activity against microorganisms significant to public health; the emphasis was on comparisons with similar bacteria. Microorganisms included:

- Escherichia coli O157: H7
- Staphyloccocus aureus
- Pseudomonas aeruginosa
- Salmonella spp.
- Campylobacter jejuni
- Listeria monocytogenes
- Shigella sonnei
- Yersinia enterocolitica
- Enterococcus hirae
- Norovirus surrogates (feline calicivirus)
- Aspergillus brasiliensis spores
- Clostridium difficile spores
- Candida albicans.

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