

Portable Chemical Sterilizer for Microbial Decontamination of Surgical Instruments, Fruits and Vegetables, and Field Feeding Equipment

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ABSTRACT

Biomedical Technologies

The Portable Chemical Sterilizer (PCS) is a revolutionary biomedical sterilization technology that provides the capability for portable, power-free, point-of-use sterilization to meet the Army's Far-Forward Surgical Teams (FSTs) needs for the rapid mobility, energy-independence, on-site sterilization of surgical instruments.

The sterilization of medical equipment is vital to prevent the risk of spreading infections or disease using contaminated surgical instruments. There were no pre-existing COTS technologies available featuring the key attributes of portability and energy-independence, and so the genesis of the development of the PCS is discussed.

The PCS chemically generates chlorine dioxide and inactivates infectious foodborne pathogens (*Escherichia coli*, *Listeria monocytogenes*, and *Staphylococcus aureus*), and tough-to-kill bacterial spores (*Bacillus stearothermophilus*, *Bacillus atrophaeus*) on the surfaces of contaminated medical equipment in 30 minutes. The technical aspects of the PCS, such as the patented chemical combination used to controllably generate the sterilant chlorine dioxide, and the key features of the patented plastic suitcase prototype are discussed in detail.

The PCS constitutes not only a major technological advance to the military – it was recognized through a 2005 Department of the

Army R&D Achievement Award for Technical Excellence, as Natick Soldier Center's Major Developmental Achievement for the 2006 Small R&D Lab of the Year Award, and publication in 7 military news journals), but it is a technology that transferred to commercial industry through 2 Patent License Agreements for use by other federal agencies (Homeland Security/Defense), State and local governments (emergency first-responders), and private industry (community hospitals, global disaster relief, Humanitarian Aid in third world countries).

The development of the PCS technology also signifies important advances in the broader scientific community through related publications in peer-reviewed journals and presentations at major international science conferences and Symposia. We discuss how the benefits of the PCS technology have also been exploited for other cross-cutting industrial applications, such as decontaminating fresh fruits and vegetables that can transmit infectious foodborne pathogens (e.g. *E. coli* on spinach, lettuce) or bio-terrorist threats through the agri-food chain. Other modifications to the chlorine dioxide chemical system led to the development of an innovative disposable handheld plastic sprayer that was designed to disinfect food handling equipment and food contact surfaces and prevent the spread of disease through secondary contamination.

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1. INTRODUCTION

The PCS is a revolutionary, patented energy-independent technology that closes a critical capability gap for the Army in the Global War on Terror.



Figure 1. Crowded conditions in an FST tent.

The PCS provides Far-forward Surgical Teams (abbreviated FSTs, see Figure 1) with the capabilities of sterilizing medical equipment on-site and without exogenous power in austere environments, to reduce the risks of spreading infections.

There were no pre-existing COTS technologies available to fill the urgent battlefield need for improved medical sterilization in far-forward areas. FSTs and Special Forces Medical detachments carry out their missions in far-forward deployments in dusty tents that are difficult to maintain as sterile environments, have limited availability of electrical power and potable water, and work in high risk areas that require rapid mobility. Contaminated instruments are useless for saving limbs and possibly lives, even in the hands of the most skilled surgeon, since using the contaminated instruments could risk spreading life-threatening infections.

1.1 Conventional Sterilization Technologies

Conventional medical sterilization involves the use of steam autoclaves to generate high temperatures ($T = 121\text{ }^{\circ}\text{C}$) at slightly elevated pressures. The traditional steam autoclaves used at Combat Support Hospitals (CSHs) are called “Berthas” (see Figures 2) that are too large (450 lbs), too difficult to maintain, and too burdensome on logistics to be transported efficiently and conveniently to these regions.



Figure 2. (Left) large “Bertha” steam autoclave at CSHs; (Right) collapsed inner chamber of “Bertha” due to overuse and limited maintenance capabilities.

1.3 Alternative Sterilization Technologies

Medical personnel in FSTs have proven that the currently available sterilization alternatives are simply inadequate for field use. Chemical sterilants either do not work well in the field (cold rinses of Cidrex or peroxyacetic acid) or are too hazardous (ethylene oxide) to be transported safely. Dental sterilizers are expensive (\$16K per unit), require electricity, and are difficult for trained medical staff to operate. Boiling contaminated instruments in water in a “lobster pot” is not sufficient to kill infectious bacterial spores, and transporting contaminated instruments with patients via aircraft to distant Combat Support Hospitals for sterilization in steam autoclaves is inefficient, expensive, time-consuming, and soon to become obsolete. “Berthas” are no longer manufactured.

2. PORTABLE CHEMICAL STERILIZER

The PCS was developed as a revolutionary, innovative, new technology to close this critical capability gap for improved medical sterilization. No other technology features all of its key attributes to overcome some of the challenges posed by “Berthas” or the Alternative Sterilization Technologies. Specifically, the PCS is a lightweight plastic suitcase (made by modifications to the COTS item) with sufficient capacity to accommodate a surgical tray and a novel chemical combination that controllably generates chlorine dioxide from a mixture of dry reagents (~200g) and water (300 mL). A standard composition kills

all contaminating vegetative cells and resistant bacterial spores within 30 minutes (Figure 3).



Figure 3. Plastic suitcase PCS prototype uses a modified COTS item and novel chemistry to sterilize surgical instrument in 30 minutes. Top left: add reagents to generate ClO_2 ; Top right: introduce tray of contaminated surgical instruments; Bottom left: close case and generate ClO_2 ; Bottom right: in 30 minutes, flush with air and remove sterile instruments – the PCS is available for immediate re-use.

Only the PCS technology is sufficiently lightweight (20 lbs.) and compact to be hand-carried and rapidly transported, uses green chemistry instead of electricity or other exogenous power sources such as fuels or open flames, functions on-site (at point-of-use) so instruments do not need to be transported via aircraft, is rugged and durable for multiple re-uses at low cost (less than \$1K per PCS and enough supplies for 30 cycles), and uses small quantities of safe, dry, easy-to-transport reagents mixed in water to generate an EPA-approved sterilant that is proven effective against a broad spectrum of infectious microorganisms. With the PCS available to them, FSTs and Special Forces Medical units can maintain a steady supply of sterile surgical instruments at the ready.

2.1 Comparison of PCS and “Bertha”

As a modern field autoclave, the PCS signifies a marked technological improvement over conventional “Bertha” steam autoclaves. The Bertha autoclaves (initial cost \$27K) weigh over 450 lbs (4-man lift), consume 9 kWatts of electricity and 5-gallons of water per cycle, occupy a 60.2 ft² cube, and can not be used on plastics. In contradistinction to the behemoth “Bertha”, the PCS is a low-maintenance, modern field autoclave designed to achieve a 95% reduction in weight, a 100% reduction in electricity usage, a 98% reduction in water consumption, safe for plastics, and a 96% reduction in cube without compromising throughput (2 PCSs or 1 “Bertha” can sterilize 4 surgical trays in 1 hour). The PCS provides FSTs and Special Forces with an improved medical sterilization technology that is truly portable, power-free, uses a proven sterilant, and functions at point-of-use to meet the high-intensity, rapid mobility demands of far forward areas.

3. CHEMICAL HEATING TECHNOLOGY

Chemical heaters (e.g, the Flameless Ration Heater) are chemical reactions that under controlled conditions release large quantities of thermal energy to heat components of MREs. In this case, the chemical basis for this heater is the iron-catalyzed magnesium-water reaction that produces potentially hazardous hydrogen gas as a product. The original PCS prototype used a thermally-insulated COTS pressure-cooker and over 23 MRE heater elements to produce large quantities of heat and steam to achieve sterilization conditions (temperatures at or exceeding 121 °C, see Figure 4).



Figure 4. Original PCS prototype was an insulated COTS pressure cooker and MRE heaters in water to effect sterilization through the production of “wet heat” (temperature above 121 °C and copious steam).

3.1 Alternative Chemical Heaters

One potential drawback of using large numbers of the MRE heater in a quasi-confined environment is the production of large quantities of potentially explosive hydrogen gas. Non-hazardous, environmentally-friendly alternative chemical heaters have been proposed to circumvent the production of hydrogen gas (Curtin et al., 2004). One candidate is the reaction of persulfate ($S_2O_8^{2-}$) and formate (HCO_2^-) in the presence of a chemical effector (AH^-). An effector is a substrate used at low concentrations that is consumed in the chemical reaction (unlike a catalyst) to initiate chemical pathways that accelerate the reaction (Table 1).

Table 1. The induced $S_2O_8^{2-}/HCO_2^-$ reaction

No.	Reaction step
1	$S_2O_8^{2-} + AH^- \rightarrow SO_4^{\bullet-} + A^{\bullet-} + SO_4^{2-} + H^+$
1'	$S_2O_8^{2-} + AH_2 \rightarrow SO_4^{\bullet-} + A^{\bullet-} + SO_4^{2-} + 2H^+$
2	$SO_4^{\bullet-} + AH^- \rightarrow A^{\bullet-} + SO_4^{2-} + H^+$
2'	$SO_4^{\bullet-} + AH_2 \rightarrow A^{\bullet-} + SO_4^{2-} + 2H^+$
3	$H^+ + A^{\bullet-} + A^{\bullet-} \rightarrow HA^- + A$
4	$SO_4^{\bullet-} + SO_4^{\bullet-} \rightarrow S_2O_8^{2-}$
5	$SO_4^{\bullet-} + A^{\bullet-} \rightarrow SO_4^{2-} + A$
6	$S_2O_8^{2-} + A^{\bullet-} \rightarrow SO_4^{\bullet-} + SO_4^{2-} + A$
7	$S_2O_8^{2-} + HCO_2^- \rightarrow SO_4^{\bullet-} + CO_2^{\bullet-} + SO_4^{2-} + H^+$
7'	$S_2O_8^{2-} + HCO_2H \rightarrow SO_4^{\bullet-} + CO_2^{\bullet-} + SO_4^{2-} + 2H^+$
8	$S_2O_8^{2-} + H_2O \rightarrow SO_4^{2-} + H_2SO_5$
9	$SO_4^{\bullet-} + HCOO^- \rightarrow SO_4^{2-} + CO_2^{\bullet-} + H^+$
9'	$SO_4^{\bullet-} + HCO_2H \rightarrow SO_4^{2-} + CO_2^{\bullet-} + 2H^+$
10	$S_2O_8^{2-} + CO_2^{\bullet-} \rightarrow CO_2 + SO_4^{\bullet-} + SO_4^{2-}$
11	$H_2SO_5 + HCOO^- \rightarrow SO_4^{2-} + CO_2 + H_2O + H^+$
12	$H_2SO_5 + HCO_2H \rightarrow SO_4^{2-} + CO_2 + H_2O + 2H^+$
13	$CO_2^{\bullet-} + CO_2^{\bullet-} \rightarrow C_2O_4^{2-}$
14	$SO_4^{\bullet-} + CO_2^{\bullet-} \rightarrow SO_4^{2-} + CO_2$

Despite its benefit to inducing the generation of heat, the effector also introduces substantial complexity to the chemical mechanism.

A similar effector was introduced into the chlorite (ClO_2^-) oxidation of sulfite (SO_3^{2-}) and had the profound effect of inducing the formation of the disinfectant chlorine dioxide (ClO_2). The formation of ClO_2 by this chemical system could kill microorganisms at much milder temperatures, and allowed subsequent PCS configurations to use lightweight, chemical-resistant plastics as construction materials. The presence of ClO_2 was confirmed using UV/Visible spectrophotometry, and the kinetics of formation of the ClO_2 was monitored using low-gate mass spectrometry (Figure 5).

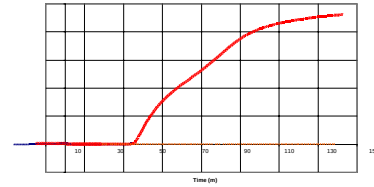


Figure 5. Chemical production of ClO_2 was determined using low-gate mass spectrometry.

A generalized reaction scheme for the production of ClO_2 by the effector-induced chlorite-sulfite chemical is presented in Table 2.

Table 2. Generalized reaction scheme for the Effector-induced chlorite-sulfite reaction.

	Reaction step
Overall	$ClO_2^- + 2SO_3^{2-} \rightarrow Cl^- + 2SO_4^{2-}$
Induction	$ClO_2^- + AH^- \rightarrow ClO^{\bullet} + ClO_2 + A + OH^-$
Propagation	$ClO^{\bullet} + SO_3^{2-} \rightarrow ClO^- + SO_3^{\bullet-}$
Propagation	$ClO_2^- + SO_3^{\bullet-} \rightarrow ClO^{\bullet} + SO_4^{2-}$
Propagation	$ClO^{\bullet} + ClO_2^- \rightarrow ClO^- + \bullet ClO_2$
Termination	$ClO^- + SO_3^{2-} \rightarrow Cl^- + SO_4^{2-}$

3.2 Early PCS Prototypes

As indicated above, the use of an alternative chemical heater that controllably generates the EPA-registered sterilant ClO_2 allowed for the use of lightweight, plastic designs for the PCS. Two of the earlier PCS configurations are shown in Figure 6.



Figure 6. Two early versions of the PCS. Top: 32-gallon tote with homemade filter unit. Bottom a 1 ft³ plastic bucket (left) before and (right) after initiating the chemical production of ClO_2 .

Results showing effectiveness of the chemical combination in a prototype for destroying vegetative borne pathogens (*E. coli* and *L. monocytogenes*) and tough-to-kill *B. stearothermophilus* spores are shown in Table 3.

Table 3. Microbial destruction in the PCS

	<u>Initial</u> <u>CFU/mL</u>	<u>Final</u>
<i>B. stearothermophilus</i> spores	1.00×10^8	0.0
<i>E. coli</i>	1.20×10^7	0.0
<i>L. monocytogenes</i>	1.34×10^8	0.0

3.3 The Plastic Suitcase PCS Prototype

As indicated in Figures 3 and 7, the plastic carry-case configuration (which was chosen by FSTs and SOCOM personnel) is the most suitable prototype for commercialization. The

plastic carry-case has sufficient capacity to hold a surgical tray of contaminated instruments.



Figure 7. The PCS plastic suitcase prototype was designed to hold an entire tray of surgical instruments (not shown).

Developing the PCS prototype from a Pelican[®] case has several advantages: Pelican cases are already used by many military units, they are lightweight, have compact dimensions, are stackable for easy storage, and can easily be modified appropriately to ensure effectiveness and the safety of the user and the environment.

A disposable reactor vessel is placed inside the suitcase to contain the chemical reagents ($\approx 200\text{g}$ mixed in 300 mL of water) that controllably produce the ClO_2 sterilant; outlet valves prevent pressure from accumulating inside the chamber during sterilization; inlet valves allow for a flow of air to remove any excess ClO_2 after the sterilization process; and filters engrafted on the outlet valves inactivate the remaining ClO_2 into benign salts that are harmless to human health and the environment.

The PCS was designed to sterilize surgical instruments contaminated with microbes in approximately 30 minutes. A tray of contaminated instruments are wrapped and placed inside the PCS. The novel chemical mixture is initiated to controllably generate ClO_2 . After 20-30 minutes of exposure, the instruments are sterilized. The remaining ClO_2 is flushed from the PCS with air for about 10 minutes and inactivated to produce benign salts by the external filters. The sterilized instruments are ready for use. The spent chemical reagents and filters can be safely disposed of. With a new set of reagents and disposable filters used for each sterilization

cycle, the PCS is ready for hundreds of re-uses on sets of contaminated instruments.

A standard chemical composition run for 45 minutes in the prototype effected the following microbial reductions:

Table 4. Representative sterilization results in the plastic carry-case Portable Chemical Sterilizer.

Reagents (g)	Water (mL)	Time (min)	Kill
181	300	30	BI
272	450	25	BI
362	600	30	BI
181	300	40	BI, HY, BT
200	300	30	BI, HY, BT

*BI designates sterilization Biological Indicators.

**HY designates sterility swab kits

***BT designates live cultures of *Bacillus stearothermophilus* spores.

Various sets of conditions were tested using biological indicators (*B. stearothermophilus* and *B. atrophaeus*) and live cultures of vegetative pathogens (*S. aureus*, *E. coli*, and *L. monocytogenes*) and tough-to-kill bacterial spores of *B. stearothermophilus*. For all of the test organisms, conditions were found that effected sterilization (6-log kill of contaminating microorganisms) using various permutations of the test variables (ClO_2 concentration and exposure time).

4. APPLICATION OF THE PCS TO FOODS AND FOOD EQUIPMENT

The basis of the sterilization process with the PCS derives from a patented chemical combination that controllably produces chlorine dioxide and environmentally-benign end-products inside a plastic suitcase prototype. The PCS technology can therefore be used in a number of alternative applications to eliminate microbes contaminating the surfaces of other types of objects or materials. We turn our attention to applications of the PCS to foods and to food handling equipment.

4.1 Application of the PCS technology to Decontaminating Fresh Fruits and Vegetables

The PCS also has applications relating to food safety, such as disinfecting fresh fruits and vegetables (Figure 8).



Figure 8. Sliced and whole tomatoes inoculated with *L. monocytogenes* and uninoculated sliced apples undergoing ClO_2 treatment inside the PCS.

The chemical combination in the PCS is adjustable to create stringent conditions of chlorine dioxide, temperature, and humidity under various test conditions to kill 10^6 *E. coli*, *L. monocytogenes*, *S. aureus*, and bacterial spores of *B. atrophaeus* and *B. stearothermophilus* and to sterilize high-grade stainless steel surgical instruments without causing chemical oxidation of the metal. The sterilization conditions can be attenuated to accommodate applications to food substrates.

Tomatoes require milder treatments to kill organisms without oxidizing the food tissue and rendering it inedible. The PCS kills (> 5-log reduction) *E. coli* and *L. monocytogenes* on inoculated, whole tomatoes without adversely affecting the color, even after 60 minutes of treatment. The interior tissue of sliced tomatoes, in comparison, tend to lose color after such prolonged exposures to ClO_2 . Conversely, fresh sliced apples did not discolor upon exposure to the ClO_2 treatments. Rather, apples retained their color, and even resisted enzymatic browning upon exposure to ambient conditions for several days.

4.2 Application of the PCS technology to Sanitizing Food Handling Equipment

As mentioned above, the PCS derives its effectiveness from a patented chemical combination that controllably produces the disinfectant ClO_2 . In addition to sterilizing contaminated surgical instruments and eliminating infectious vegetative pathogens from fresh fruits and vegetables, ClO_2 can also be used to sanitize food handling equipment and food contact surfaces (counter tops, cutting boards) to prevent the spread of foodborne illnesses through secondary contamination. In these instances, the contaminated objects can not be placed inside the PCS for exposure to ClO_2 treatments, and so an alternative method needs to be developed to deliver the ClO_2 to the contaminated surfaces.

One possible solution is generating the ClO_2 inside a collapsible, disposable handheld plastic sprayer (see Figure 9). The plastic sprayers are available commercially and have a volume exceeding 900 mL. As fluid is added to the plastic pouch, a gusseted bottom opens up and allows the sprayer to stand up independently. A dilute solution of ClO_2 can be generated this way in the morning and last an entire day without decomposing and losing its anti-microbial effectiveness.



Figure 9. The ClO_2 -generating capacity of the PCS technology can be exploited to produce a disinfectant solution in a collapsible, disposable handheld sprayer.

The sprayer can be used to sanitize field feeding equipment and food contact surfaces that can harbor infectious pathogens due to either poor hygiene or contact with contaminated foods. A sprayer that is easy to use, effective at low doses against microbes, and convenient can be used to disinfect food contact surfaces and reduce the potential for spreading foodborne illnesses through secondary contamination. Figure 10 demonstrates how the plastic sprayer can easily be handled to spray

and wipe cutting boards and other food contact surfaces to quickly and efficiently eliminate microbes from these surfaces. In this case, with the handheld sprayer, the ClO_2 concentration is relatively dilute and the contact time with the surfaces is fairly short (10-30 seconds), and benefits of the ClO_2 sprayer are not contravened by potential oxidative damage to these surfaces.



Figure 10. Simply spray-and-wipe with the handheld ClO_2 sprayer to conveniently disinfectant food contact surfaces. The chemistry for generating ClO_2 is the same as that used to decontaminate surgical instruments with the PCS.

The novel chemical combination that was used to generate ClO_2 and decontaminate surgical instruments with the PCS suitcase prototype can also be used: 1) to eliminate microbes and potential bio-terrorist threats in the agri-food chain by treating fresh fruits and vegetables; and 2) to generate ClO_2 in a handheld plastic sprayer to conveniently disinfect field feeding equipment and food contact surfaces to improve hygiene and prevent the spread of illness through secondary contamination.

5. SUMMARY

The PCS is a revolutionary technology that provides portable, energy-independent, on-site sterilization of surgical instruments to meet an urgent battlefield for Army's FSTs and Special Forces Medical units by closing a critical capability gap for improved medical sterilization technologies. The PCS and the underlying chemistry used to produce the putative sterilant ClO_2 are the objects of two separate patents that were generated by federal funding by a collaboration of two Army agencies: the Natick Soldier Center and the

Institute of Surgical Research. The PCS technology was transferred to private industry via two separate Patent License Agreements (PLAs) with two different small businesses. The transfer of the PCS technology to commercial industry through these PLAs was accomplished under the guidance of DoD TechLink. As a licensed patent, the commercial version of the PCS technology will be available to other federal agencies in Homeland Security/Defense, emergency first-responders at the State and local government levels, and to the private sector. Private companies will be able to market the PCS technology as emergency back-up units for use in community hospitals, where recent autoclave failures have led to deaths and costly law suits. Licensing the PCS technology will also increase the competitiveness of US companies in global markets by providing them with a portable, energy-independent medical technology that can be marketed to global disaster relief organizations (Katrina, tsunamis) and to organizations involved in providing Humanitarian Aid to third world countries (UNICEF, Doctors without Borders).

Further developments of this technology will enhance the military's capabilities for decontaminating bio-threats that cause foodborne illnesses, and ensure the health and safety of deployed Warfighters and the Future Force. As recent outbreaks of *E. coli* in spinach

and lettuce in the US consumer market have demonstrated, the agri-food chain is vulnerable to the rapid promulgation of bio-terrorist threats. Present industrial practices for washing and disinfecting fresh fruits and vegetables are simply inadequate for eliminating these health threats. There is a need for improved intervention technologies to increase the safety of fresh produce and to eliminate possible agri-terrorist threats both in the domestic US consumer market and in the military supply chain. Global military deployments that purchase produce from local growers who might use contaminated soils, irrigation water, or fertilizers; who might not use pesticides, and who might exercise unsanitary or unhygienic farming practices need improved disinfection technologies to counter emerging or more virulent bio-threats endemic to those regions. Future work will focus on optimizing the PCS technology for use on fresh fruits and vegetables to ensure the safety of the consumer.

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