

Efficacy of decontamination and sterilization of a single-use single-incision laparoscopic surgery port

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Objective—To determine the efficacy of decontamination and sterilization of a disposable port intended for use during single-incision laparoscopy.

Sample—5 material samples obtained from each of 3 laparoscopic surgery ports.

Procedures—Ports were assigned to undergo decontamination and ethylene oxide sterilization without bacterial inoculation (negative control port), with bacterial inoculation (*Staphylococcus aureus*, *Escherichia coli*, and *Mycobacterium fortuitum*) and without decontamination and sterilization (positive control port), or with bacterial inoculation followed by decontamination and ethylene oxide sterilization (treated port). Each port underwent testing 5 times; during each time, a sample of the foam portion of each port was obtained and bacteriologic culture testing was performed. Bacteriologic culture scores were determined for each port sample.

Results—None of the treated port samples had positive bacteriologic culture results. All 5 positive control port samples had positive bacteriologic culture results. One negative control port sample had positive bacteriologic culture results; a spore-forming *Bacillus* sp organism was cultured from that port sample, which was thought to be an environmental contaminant. Bacteriologic culture scores for the treated port samples were significantly lower than those for the positive control port samples. Bacteriologic culture scores for the treated port samples were not significantly different from those for negative control port samples.

Conclusions and Clinical Relevance—Results of this study indicated standard procedures for decontamination and sterilization of a single-use port intended for use during single-incision laparoscopic surgery were effective for elimination of inoculated bacteria. Reuse of this port may be safe for laparoscopic surgery of animals. (*Am J Vet Res* 2013;74:934–938)

Decontamination and sterilization of laparoscopic SUDs for humans undergoing MIS is a controversial method.^{1–4} Such devices are typically reused because of economic concerns; however, reuse of disposable instruments may increase risk of infection for patients.^{1–4} Single-use surgical devices are typically made of less robust materials (eg, plastic and rubber) than are nondisposable surgical devices; therefore, decontamination and sterilization of SUDs can be difficult, and such devices are prone to mechanical malfunction when reused.^{1,4}

ABBREVIATIONS	
MIS	Minimally invasive surgery
SUD	Single-use device
TSB	Tryptic soy broth

Reuse of SUDs is common in the field of veterinary surgery. There are few reports of mechanical failure or adverse effects for animals when SUDs are reused for performance of surgery. To the authors' knowledge, it is unknown whether this scarcity of information is attributable to a lack of such effects or to underreporting of adverse events.

Veterinarians typically reuse laparoscopic SUDs because of the high cost of such devices. Veterinary surgeons and surgeons performing procedures for humans in developing or poor countries² may have justification for reuse of SUDs during MIS because of economic concerns. However, adverse effects of such reuse of devices may be different between humans and other animals.

A disposable instrument port has been developed for use during single-incision laparoscopic surgery of

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Laparoscopic surgery ports were provided by Covidien.

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humans. Although the material composition of that port is proprietary information of the manufacturer, the device has a gross appearance similar to firm, malleable foam. The advantage of this port for the performance of laparoscopic surgery is that 3 to 4 instruments can simultaneously be used through a single short incision. Results of other studies^{3,a} indicate the device is useful for MIS in dogs and cats. However, the cost of the device is approximately \$250 to \$400; therefore, use of the device by veterinarians for single-incision laparoscopy of animals may be economically impractical.

The purpose of the study reported here was to determine the efficacy of decontamination and sterilization of a disposable port intended for use during single-incision laparoscopy. We hypothesized that a commonly used method of surgical instrument decontamination and sterilization would be efficacious for elimination of bacteria on this laparoscopic instrument port.

Materials and Methods

Sample—A disposable instrument port^b intended for use during single-incision laparoscopy was used in this study (Figure 1). A power analysis was performed to determine the sample size of laparoscopic ports to be tested during the study. This calculation was performed with an α value of 0.05, a β value of 0.20, a prevalence of positive bacteriologic culture results of 95% for a positive control port group, a prevalence of positive bacteriologic culture results of 5% for negative control and treated port groups, and a null difference value of 0.1. A sample size of 5 was determined for each of the 3 tested ports.

Procedures—Three ports underwent testing 5 times each during this study. Prior to the start of the study, each of the 3 ports was randomly assigned to undergo negative control, positive control, or treated group procedures as determined by drawing numbers from an opaque bag. The assigned procedure for each port remained the same for the duration of the study. For each of the 5 testing times in the study, samples of ports were obtained for bacteriologic culture by use of a sterile 6-mm biopsy punch.^c All samples of ports were obtained from the central concave region of the ports. Samples of plastic cannulas and insufflation tubing of ports were not obtained for testing. The positive control and treated ports were inoculated with *Escherichia coli*, *Staphylococcus aureus*, and *Mycobacterium fortuitum* during each testing time.

The negative control port was not inoculated with bacteria. For each testing time, that port was placed in sterilization wrap and sterilized in ethylene oxide^d immediately following collection of each port material sample. Port material samples were placed in 5 mL of sterile TSB^e and incubated at $95 \pm 2\%$ relative humidity in 5% to 10% CO₂ at 35° to 37°C. Bacteriologic cultures of port material samples were examined for growth at 18 to 24 hours after the start of incubation and again at ≥ 48 hours after that time. The ports and port material samples were handled by use of an aseptic technique, including the use of sterile single-use biopsy punches, blades, and latex surgical gloves.

The positive control port was exposed to *E coli* American Type Culture Collection No. 25922, *S aureus*

American Type Culture Collection No. 29123, and *M fortuitum* (isolated from a clinical patient and identified via DNA sequence analysis). Each bacterial isolate was incubated separately in TSB at $95 \pm 2\%$ relative humidity in 5% to 10% CO₂ at 35° to 37°C. The bacteria were combined and suspended in sterile deionized water with a cell density of approximately 1.5×10^5 CFUs/mL of each species. The positive control port was submerged in the bacterial suspension for 30 minutes at approximately 21°C, then port material samples were collected with a sterile biopsy punch. Port material samples obtained at each of the 5 testing times were placed in 5 mL of sterile TSB^e and incubated at $95 \pm 2\%$ relative humidity in 5% to 10% CO₂ at 35° to 37°C. Bacteriologic cultures of positive control port material samples were examined 18 to 24 hours after the start of incubation to detect *E coli* and *S aureus* growth and from 48 hours to 1 week after that time to detect *M fortuitum* growth.

The treated port was inoculated with bacteria via the same method used to inoculate the positive control port. The port was then decontaminated via rinsing with tap water for 1 minute and soaking in an enzymatic cleaner (dilution, 3:100)^f and brushing with a sponge scrub brush and pipe cleaner brush for 5 minutes. The port was then rinsed with tap water, dried with compressed air, and packaged and heat-sealed in plastic wrap. The wrapped port was then exposed to ethylene oxide for 16 hours via a standard protocol^g at 50°C and $> 30\%$ relative humidity (as determined with a humidity chip^g). Sterilization indicators^h were used to ensure sterile conditions were attained during each ethylene oxide sterilization cycle. After sterilization, a port material sample was collected and aerobic bacteriologic culture was performed via the same method used for negative and positive control ports.

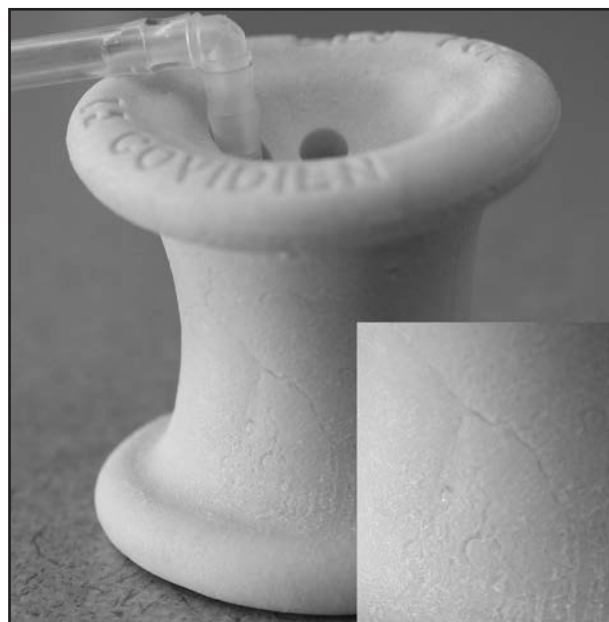


Figure 1—Photograph of the foam portion of a single-use port intended for use during single-incision laparoscopy. Notice the concave central area of the port and the porous appearance of the foam (inset). Rigid cannulas for this device are not shown.

Each incubated TSB tube of port material samples was visually inspected for growth (turbidity) and sub-cultured onto Columbia base with 5% sheep blood,ⁱ Columbia base with 5% sheep blood and colistin and nalidixic acid,^j and MacConkey agar^k to confirm the presence or absence of each species of bacteria. Plates were incubated at 95 ± 2% relative humidity in 5% to 10% CO₂ at 35° to 37°C for 18 to 24 hours for detection of *E coli*² and *S aureus* and for ≥ 48 hours for detection of *M fortuitum*.

Statistical analysis—For each species of bacteria detected for each port material sample, an ordinal score was assigned. Scores assigned for each port material sample included 0 (no bacteria detected), 1 (1 species of bacteria detected), 2 (2 species of bacteria detected), or 3 (3 species of bacteria detected). Data were expressed as median and range values. A Wilcoxon signed rank test and a Kruskal-Wallis rank sum test with χ^2 approximation were used to determine statistical differences in bacterial culture scores among ports. Values of $P < 0.05$ were considered significant. Statistical analysis was performed by use of software.¹

Results

Significant ($P = 0.003$) differences in bacterial culture scores were detected among groups of port material samples. Bacteriologic culture scores for the positive control port material samples (median score, 3 [range, 1 to 3]) were significantly ($P = 0.010$) higher than those for the negative control port material samples (median score, 0 [range, 0 to 1]). For the negative control port sample obtained during the first test, bacteriologic culture results were positive for a spore-forming *Bacillus* sp. The positive control port material sample obtained during that time had positive bacteriologic culture results for all 3 species of inoculated bacteria, and the treated port material sample obtained during that time had negative bacteriologic culture results. None of the treated port material samples collected during the study had positive bacteriologic culture results. A significant

Table 1—Proportion of negative control (n = 5), positive control (5), and treated (5) laparoscopic surgery port samples with positive bacteriologic culture results for *Staphylococcus aureus*, *Escherichia coli*, and *Mycobacterium fortuitum* after inoculation with those bacteria.*

Test group	Bacterial species		
	<i>E coli</i>	<i>S aureus</i>	<i>M fortuitum</i>
Negative†	0/5	0/5	0/5
Positive	4/5	5/5	4/5
Treated	0/5	0/5	0/5

Data are number of laparoscopic surgery port samples with positive bacteriologic culture results for a bacterial species/total number of port samples in the test group.

*Laparoscopic ports were not exposed to bacteria (negative control port samples); were exposed to 1×10^5 CFUs/mL of *S aureus*, *E coli*, and *M fortuitum* for 30 minutes (positive control port samples); or were exposed to 1×10^5 CFUs/mL of *S aureus*, *E coli*, and *M fortuitum* for 30 minutes, then cleaned and sterilized with ethylene oxide (treated port samples). †Bacteriologic culture results for 1 negative control port sample were positive for a *Bacillus* sp organism; data for this organism are not included because ports were not inoculated with that species of bacteria and it was thought to be an environmental contaminant.

difference in bacterial growth score was detected between the treated port material samples (median, 0 [range, 0 to 0]) and positive control port material samples ($P = 0.006$) but not between the treated port material samples and negative control port material samples ($P = 0.42$). Data regarding bacteriologic culture results for ports were summarized (Table 1).

Discussion

Results of the present study indicated decontamination and sterilization of the multichannel laparoscopy port were effective. These findings suggested that this port may be reused for the performance of MIS in animals. However, further studies are indicated to determine whether reuse of the device would cause complications such as infection in animals or equipment malfunction.

The multichannel single-use laparoscopy port used in this study is a unique peritoneal access device because it permits simultaneous passage of 3 to 4 cannulas through a single 20- to 30-mm incision.^{5,a} This method may have advantages over traditional laparoscopic techniques, which require creation of an incision for each instrument. Among the potential benefits of a single-incision laparoscopic technique are creation of a small incision and minimization of the number of incisions created and instrument cannulas used during a procedure.^{5,a} Use of techniques in which the number and size of incisions are reduced is becoming common for MIS in humans^{7,8} and other animals.^{5,9,10,a} In the authors' experience, the port used in the present study has been useful for the performance of laparoscopic-assisted gastrointestinal tract exploratory surgery in animals. Furthermore, for small patients, the port allows exteriorization of organs (such as portions of the intestinal tract) for the performance of extracorporeal procedures without increasing the size of the original incision. This technique may decrease morbidity of animals undergoing MIS, compared with that for animals undergoing traditional open surgical techniques.⁷⁻¹² Therefore, use of this port by veterinarians may become more common for performance of MIS in animals. However, the port has a high cost (\$250 to \$400). Because this port is labeled as an SUD, decontamination and sterilization of the device are not advised by the manufacturer. However, results of the present study suggested that decontamination and sterilization of the device may be effective for reduction of the number of bacteria, which may allow safe reuse of the port for laparoscopic procedures in animals.

Reuse of SUDs for laparoscopy of humans is controversial. Because few complications of reuse of such devices have been detected and the devices are expensive, reuse of SUDs may be justifiable.^{2,13} Results of other studies^{2,13} indicate adverse clinical sequela of reuse of SUDs have not been detected and the cost of surgical materials for open and laparoscopic surgical procedures for humans may be substantially (> \$3,000) different. However, total costs (including hospitalization costs) of open and laparoscopic surgical procedures for humans may be similar or such costs for laparoscopic procedures may be less than those for open surgical procedures.¹³ Findings for veterinary patients may be similar.

Reuse of SUDs has potential disadvantages. Patients undergoing surgical procedures in which SUDs are reused may have higher risk of infection versus patients undergoing surgical procedures in which such devices are not reused because of incomplete decontamination of SUDs or injury attributable to mechanical failure of such devices.¹⁻⁴ Results of other studies^{3,4} indicate incomplete removal of organic material (determined via scanning electron microscopy and radionuclide labeling techniques) in up to 100% of contaminated SUDs that have been cleaned. Although low amounts of residual organic material were found on SUDs in those studies,^{3,4} appropriate disinfection for killing of microbial organisms was achieved. Determination of the clinical relevance of the results of those studies may be difficult because multiple factors may affect such findings, including type of device used, amount of prior use of a device, species of animal undergoing surgical procedures, and methods used for decontamination and sterilization of a device. Only a small amount of information is available regarding clinical complications associated with reuse of SUDs in animals undergoing surgery. Other authors¹⁴ estimated that infection is caused by contaminated instruments in only 1 of 1.8 million gastrointestinal procedures for humans. Further studies may be indicated to determine the risks and complications associated with reuse of SUDs for animals undergoing surgical procedures.

In the present study, we attempted to determine the possibility of transmission of infectious bacteria to animals via a reused multichannel laparoscopy port by assessing the efficacy of decontamination and sterilization techniques that are typically used for surgical instruments. This was thought to be important information because of the malleable and porous properties of the ports. The species of bacteria used for inoculation of ports in the present study were selected because they were thought to be representative of various bacteria that typically cause contamination of instruments and infection of animals after surgery.¹⁵ The concentration of each species of bacteria used to inoculate ports (1.5×10^5 CFUs/mL) in the study reported here was intended to simulate instrument contamination severe enough to cause infection in animals. A *Mycobacterium* sp was included because the response of organisms of this genus to disinfectants is different from that of *Staphylococcus* spp and organisms of the family Enterobacteriaceae, and this organism is a reported cause of infection in humans that undergo laparoscopy in countries where sterilization and reuse of SUDs is commonly performed.¹⁶ The technique used to decontaminate and sterilize laparoscopic ports in the present study was the same technique used in our hospital for cleaning and sterilization of similar surgical materials. Results of this study indicated bacteria were not detected in treated port material samples after decontamination and sterilization. Thus, the decontamination and sterilization protocol used in the present study may be appropriate for preparation of SUDs for reuse in a clinical setting. Further studies of this decontamination and sterilization technique for SUDs used during clinical procedures of animals are warranted.

Several limitations of the present study were identified. The ports were not tested to detect viruses, fungi,

or protozoa. Although transmission of viruses among human patients is an important concern, it may not be as important for dogs undergoing surgery because viral diseases of dogs may have lower prevalence and virulence than those of humans. However, transmission of some viral pathogens to dogs undergoing surgery, such as hepatitis C virus, may be an important concern.¹⁷ In addition, FIV and FeLV may be transmitted on instruments between cats undergoing surgery. Postoperative fungal infections in humans (especially infections caused by *Candida* spp organisms) are increasing in prevalence, and further studies of disinfection techniques for fungi are warranted.¹⁸ Another limitation of the present study was that laparoscopic ports were not exposed to blood or other biological fluids of animals; exposure of ports to such fluids might alter the efficacy of cleaning and sterilization. Additionally, none of the ports were subjected to mechanical trauma and each port was tested only 5 times. The structural integrity of the ports, and therefore the ability to effectively decontaminate and sterilize them, may be altered with repeated use; however, this was not evaluated in the present study and conclusions regarding that possibility could not be determined. Also, only the foam portions of laparoscopic ports were tested via bacteriologic culture. Plastic cannulas and insufflation tubing of ports were not tested. We did not test those portions of the ports because we thought that these rigid structures would be less likely to harbor bacteria after decontamination and sterilization versus the foam portions of the ports.

Results of this study suggested that decontamination and sterilization were effective in eliminating bacterial viability in vitro on laparoscopic ports intended for single use. These results may support reuse of this SUD for MIS of animals. Reuse of such laparoscopic ports may be safe for animals and economically beneficial for veterinarians. However, further in vitro and clinical testing is warranted to identify potential complications of reuse of this device, such as infection in animals or port malfunction, before routine reuse can be recommended.

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- e. TSB, Hardy Diagnostics, Santa Maria, Calif.
- f. Bio-zyme, Osceola Supply, Tallahassee, Fla.
- g. Humidichip, Andersen Products, Haw River, NC.
- h. Ethylene oxide gas dosimeter, Andersen Products, Haw River, NC.
- i. CBA, Hardy Diagnostics, Santa Maria, Calif.
- j. CNA, Hardy Diagnostics, Santa Maria, Calif.
- k. MAC, Hardy Diagnostics, Santa Maria, Calif.
- l. JMP, version 9.0, SAS Institute Inc, Cary, NC.

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