TAB 10 **FDA Reviewer's Literature Survey to Determine Correlation of Infection Rates to Microbiological Endpoints:**

- **Healthcare Personnel Handwashes – Colleen Rogers, Ph.D.**
- **Surgical Hand Scrubs – Michelle M. Jackson, Ph.D.**
- **Patient Preoperative Skin Preparations – Peter Kim, M.D.**

Report of Medical Officer Consultation

Requesting Division:	Division of Over-the-Counter Drug Products, HFD-560
Contact:	Debbie Lumpkins Microbiology Team Leader Phone: 301-827-2222
Date of MO's Review:	6/24/04
Drug Indication:	Patient Preoperative Skin Preparation
Re:	Use of surrogate microbiologic endpoint criteria described in the FDA Proposed Tentative Final Monograph (TFM) for Health Care Antiseptic Drug Products, Effectiveness Testing of a Patient Preoperative Skin Preparation (published in the Federal Register on June 17, 1994) for demonstration of effectiveness for the Patient Preoperative Skin Preparation indication.

I. Background

Many factors influence surgical site infections (SSIs). These factors include: type of surgery (clean, clean-contaminated, etc.), length of procedure, and environment in which surgery occurs. Healthcare providers attempt to reduce the incidence of SSIs by altering the surgical environment (e.g., regulating airflow in the surgical suite, sterilizing equipment, etc.), maintaining strict adherence to surgical hand scrub technique, and through use of sterile gowns, gloves, and drapes. In addition, patients' surgical sites are prepared with various antimicrobial products to remove transient populations of bacteria from the skin. Peri-operative systemic antibacterial therapy may also reduce endogenous populations of bacteria that seed the surgical site and bloodstream of the patient.

The Food and Drug Administration (FDA) currently approves products indicated for the patient preoperative skin preparation indication based on a surrogate microbiologic endpoint of log reductions in bacteria cultured from "dry" and "wet" anatomic sites on healthy volunteer subjects.[14] However, it is not clear that the log reductions required in the Tentative Final Monograph (TFM) result in clinical benefit to patients (that is, reduction in SSI).

Recently, clinical reviewers in the Division of Anti-Infective Drug Products searched the literature for evidence of a direct link between bacterial log reduction and decreased incidence in SSIs. They searched for articles in the National Library of Congress' PubMed system and employed the following search criteria: "skin," "surgery," "infection," AND "prevention." The reviewers also pulled and reviewed pertinent references from the bibliographies of key articles. The review staff screened over 400 articles (see Appendix 1 for complete listing) and attempted to perform a meta-analysis to

determine the clinical validity of microbiologic endpoints described in the TFM for the patient preoperative skin preparation indication. Criteria for inclusion in the meta-analysis were: (1) bacterial log reduction data available pre-scrub and at minimum post-scrub and (2) definition of SSI and rate of SSI during follow-up reported in the reference.

The reviewers concluded that inadequate information existed in the literature to perform a meta-analysis. The majority of the studies that met the inclusion criteria were performed in animals. No human or animal studies found an association between rate of SSIs and number of colony forming units (CFUs) of bacteria found on either the pre- or post-scrub cultures.

During the review of published literature, the reviewers also sought to determine if evidence existed that would support the use of alternate microbiologic criteria that were predictive of clinical benefit. A number of references, in which an association was found between levels of endogenous bacteria and infection incidence in wounds, were reviewed. Authors of these studies have proposed that wounds contaminated with less than 10^5 CFUs per gram of tissue or per milliliter (ml) of fluid are much less likely to become infected than those contaminated with greater than 10^5 CFUs. This theory (referred to as the "10^5 theory" in the balance of this review) has proven useful in the assessment of potential infection in burn wounds, skin flap survival, battlefield wounds, and civilian traumatic wounds. However, exceptions to this "rule" exist.[19,22]

II. Summary of Literature Review

SSI PREVENTION, "STATE OF THE ART"

An estimated 27 million surgical procedures are performed annually in the United States.[1] Based on data from the CDC's National Nosocomial Infection Surveillance (NNIS) system, SSIs are the third most frequently reported nosocomial infection, accounting for 14-16% of all nosocomial infections among hospitalized patients.[12] Several studies corroborate that increased length of stay and increased hospital care costs are associated with SSIs.[7,31] Advances in infection control practices include improved operating room ventilation and sterilization methods; use of sterile gowns, gloves and drapes; refined surgical technique; and the availability of improved antimicrobial prophylaxis. Risks associated with SSIs include characteristics of the patient, type and duration of operation, surgical personnel, and hospital.[27]

Quantitatively, Krizek et al. have observed that if a wound is contaminated with $> 10^5$ microorganisms per gram (gm) of tissue or milliliter (ml) of body fluid, the risk of developing a SSI post closure is markedly increased.[22] However, the dose of contaminating organisms required to produce infection may be much lower when foreign material is present at the site. Investigators have observed that an innoculum of as little as 100 staphylococci per gram of tissue introduced on silk sutures may cause infections in mice and humans.[11,20,29] For most SSIs, the source of pathogens is the endogenous flora of the patient's skin, mucous membranes, or hollow viscera.[2] Exposed tissues are at risk

for contamination with endogenous flora when mucous membranes and skin are incised.[19] Exogenous sources of SSI pathogens include surgical personnel, the operating room environment (including air), and all tools, instruments, and materials brought to the surgical field during an operation.[5,9,15,24,28]

Measures to control patient-specific sources of SSI pathogens include preoperative showering, shaving, and skin preparation. Though preoperative antiseptic showering decreases skin microbial colony counts,[17] it has not definitely been shown to reduce SSI rates.[3] Literature suggests that preoperative hair removal increases SSI rates.[39] The relative contribution to SSI incidence by different methods of hair removal remains controversial. According to the Guideline for Prevention of Surgical Site Infection written by the Hospital Infection Control Practices Advisory Committee (HICPAC) and published in 1999, "No studies have adequately assessed the comparative effects of the preoperative skin antiseptics on SSI risk in well-controlled, operation specific studies."[27]

STUDIES ATTEMPTING TO ANSWER THE QUESTION OF CLINICAL RELEVANCE: ARE REDUCTIONS IN MICROBIAL COUNTS ON THE SKIN OBTAINED WITH PATIENT PREOPERATIVE SKIN PREPARATIONS ASSOCIATED WITH A REDUCTION IN THE INCIDENCE OF SSIs?

In a study by Cronquist et al., 608 human neurosurgical patients were studied between January 1999 and May 2000 at Columbia Presbyterian Medical Center. The purpose of the study was to describe the density of bacterial counts on the skin with pre- and post-scrub cultures of neurosurgical patients and examine the association between total bacterial counts [in colony forming units (CFUs) converted to log scale] at the operative site and the incidence of surgical site infections (SSIs). The authors used National Nosocomial Infection Surveillance (NNIS) criteria to define SSIs. Cultures were taken using a sterile swab to sample a quarter-sized area of skin. Sampling was performed in a circular motion applying the pressure used when manipulating a pencil eraser. All patients were followed for at least 60 days postoperatively. Out of the 608 neurosurgical patients followed, 20 developed SSIs. According to the authors, the majority (89%) of surgeries were prepared in the following manner: hair was clipped; the surgical site was scrubbed with povidone iodine, rinsed with isopropyl alcohol, and then painted with povidone iodine. Perioperative antibiotics were administered to all patients. The authors found no correlation between SSIs and high CFUs (any bacteria on culture) versus low CFUs (no bacteria on culture) on post-scrub cultures taken after application of antiseptic and just before the surgeon draped the area for incision. The authors also found no correlation between infection and high CFUs ($> 10^5$ log CFU) versus low CFUs ($\leq 10^5$ CFU) on pre-scrub cultures taken after hair removal but before application of the antiseptic agent. The mean log CFUs for pre-preparation samples taken from the head (craniotomies and VP shunt insertions) and back were 4.13 log and 2.39 log, respectively. The mean log CFUs for post-preparation samples taken from the head and back were 0.62 and 0.54, respectively. Coagulase-negative staphylococci were the most prevalent organisms isolated in both pre- and post-preparation cultures.[6]

Georgiade et al. studied 150 human patients (reportedly 146 evaluable for efficacy) undergoing elective surgery. This was an open study designed to assess the bactericidal and clinical efficacy of a preoperative skin preparation procedure: 7.5% povidone iodine surgical scrub followed by 10% povidone iodine antiseptic solution. Cultures were obtained using a scrub cup technique. No infection criteria were specified. The authors stated that patients were "evaluated for presence of infection at the incision and suture sites during the 10-12 days following surgery..." Of 99 patients with bacterial colonization of skin prior to surgery, 84 patients (85%) had no detectable bacteria at the completion of surgery. Bacterial flora persisted after surgery in the remaining 15 patients (15%). Despite this, no SSIs were reported in any of the patients.[18]

Desrochers et al. compared two different preoperative skin preparations in 117 cattle. The study was performed at Kansas State University veterinary teaching hospital between September 1993 and August 1994. Group 1 (PI Group) consisted of 60 animals prepared with 0.75% povidone iodine soap and 70% isopropyl alcohol. Group 2 (CHG Group) consisted of 57 animals prepared with 4% chlorhexidine gluconate and 70% isopropyl alcohol. The purpose of the study was to determine: (1) which preparation produced lower CFUs on post-preparation cultures and (2) if a greater reduction in CFUs translated into a lower SSI rate. Animals were administered antibiotics before or shortly after the surgery. Cultures were performed using RODAC (replicating organism detection and counting) plates applied using gentle pressure to the skin surface at the proposed sites of incision. Surgical wound infection was defined as, "inflammation and purulent discharge from the incision or by culture of organisms from fluid obtained from the wound." Surgical wounds were evaluated daily for hospitalized cattle and then 14 to 30 days after surgery by either the surgeon or by telephone with the referring veterinarian or owner. Suture abscesses were not considered SSIs as per NNIS criteria. The authors note certain peculiarities associated with bovine surgery when compared with human surgery. The operative site is often larger. The animal is often conscious and standing. Surgical procedures are often performed in a contaminated surgical theater. The CHG Group had significantly fewer CFUs (mean CFUs=2.79) and a greater percent reduction in CFUs (98.6%) post-preparation as compared with the PI Group (mean CFUs=10.27), 93.3% reduction, $p<0.005$. However, the wound infection frequency for clean surgical procedures was not significantly different between the PI Group and the CHG Group, 9.8% and 10.7%, respectively.[8]

Rochat et al. studied 60 canine patients undergoing ovariohysterectomy divided two skin preparation techniques. Thirty dogs received a 2-step preparation: Povidone iodine (0.75% available iodine) soap was used for a non-sterile scrub. The scrub was then repeated using sterile materials. The proposed surgical site was then sprayed with 90% isopropyl alcohol followed by povidone iodine solution (1% iodine). Thirty dogs received a 1-step preparation: Duraprep (nonsurfactant iodophor polymer complex (0.5% iodine) combined with isopropyl alcohol (70% W/W). Two sets of cultures were obtained using RODAC plates: (1) after the proposed surgical site was clipped and vacuumed, but before the antiseptic was applied, and (2) after the proposed surgical site was prepared. Surgical sites were evaluated for infection the day after surgery and at the time of suture removal (7-10 days later). Infection was defined by observation of

"inflammation and purulent discharge." Though the authors reported no SSIs, they did note the following complications: suture abscesses in three dogs (two in the 2-step group, one in the 1-step group), swelling and serosanguinous discharge in one dog (2-step), and a swollen incision line and mild pyrexia in one dog (1-step). None of the dogs that developed post-operative complications had growth on post-preparation cultures. The authors reported no significant difference in the percent reduction of microbial numbers after skin preparation for either group. Actual numbers were not reported.[35]

Galuppo et al. studied 53 horses undergoing exploratory celiotomy for colic. The horses were prepared using one of two techniques. In Group 1, 26 horses were prepared using povidone iodine and alcohol. In Group 2, horses were prepared with a film-forming iodophor complex. Antibiotics were administered to each horse 30 minutes to 1 hour prior to induction of anesthesia. Continuation of antibiotics depended on the clinical disorder. Quantitative cultures were performed using RODAC plates and swabbing a 4 x 4 cm area of skin. Cultures were taken before and after skin preparation, following skin closure, and after recovery from general anesthesia. Infection was defined as "any evidence of incisional drainage" from a surgical wound. Wounds were evaluated daily while in hospital and follow-up information was obtained from owners and referring veterinarians by telephone. The authors found that both skin preparation techniques were equally effective in reducing CFUs by 99%. They observed incisional drainage in 14 of 53 horses (Group 1: 8 of 26, Group 2: 6 of 27). In Group 1, post-preparation mean CFUs between horses with and without drainage were 0.3 and 0.6, respectively. In Group 2, post-preparation mean CFUs among horses with and without drainage were 9.2 and 0.9, respectively.[16]

Stubbs et al. studied 100 canine patients undergoing orthopedic, neurological, and soft tissue procedures randomly divided into two skin preparation techniques at the University of Florida, Veterinary Medical Teaching Hospital. Forty-one dogs were prepared with 4% chlorhexidine gluconate (CHG). Fifty-six dogs were prepared with 3% chloroxylenol (PCMX). Quantitative cultures were performed using RODAC and swab cultures after hair removal, after surgical scrub, and immediately postoperatively. Surgical wounds were examined within 24 hours of surgery and again after suture removal. Criteria used to identify wound infection included "a purulent exudate or cardinal signs of inflammation, or both." Bacterial counts after surgical preparation were significantly lower in the CHG group compared with the PCMX group ($P < .0001$). However, the incidence of wound infections did not differ significantly between the PCMX and CHG groups: 2/54 (3.7%) and 2/39 (5.1%), respectively. The authors noted that none of the dogs that had SSIs had unusually high CFU counts relative to the dogs that did not develop SSIs.[37]

LITERATURE ON THE "10^5 THEORY"

Krizek and Robson state that a surgical site is not germ-free, even in the absence of infection. A delicate balance exists that allows humans to survive in the presence of a multitude of microbes. When no infection is present, equilibrium exists between factors

of host resistance and the actions of bacteria. Disequilibrium may occur when either host defense mechanisms are impaired or there is an increase in the bacterial innoculum. However, the mere presence of organisms in a wound is less important than the level of bacterial growth. For most bacteria, the level of growth necessary to cause wound infection and sepsis is greater than 100,000 (or 10^5) bacteria per gram of tissue.[22] Only β-hemolytic streptococci appear capable of routinely causing infection at levels less than 100,000 bacteria/gram of tissue.[33] Foreign bodies, such as suture and drains, may also decrease host resistance and promote an environment susceptible to infection.[34] Rodeheaver et al. demonstrated that as little as 5 milligrams (mg) of soil reduced the number of bacteria necessary to cause wound infection in an animal model from 1 million to 100.[36] In 1956, Elek demonstrated in human volunteers that the presence of a single silk suture decreased the number of staphylococci necessary to form a pustule from greater than 10^6 to 10^2, a 10,000 fold decrease.[10]

According to Krizek and Robson, World War I French physicians made the first correlation of a quantity of bacteria affecting the risk of infection. If a battlefield wound was more than 15 hours old, the wound was debrided, cultured, irrigated with Dakin's solution, and packed with gauze. The patient was evacuated to the base hospital along with the culture plate. If no streptococci grew on the plate or less than five colonies of any other organism grew, then the packing was removed and the wound was closed. If any streptococci or greater than five colonies of any other bacteria grew, then the wound was left open to heal by secondary intention. Krizek and Robson note that this early wound treatment not only recognized species differences in virulence, but was also the first documentation of quantitative evaluation of the bacterial contamination of a wound.[22]

Additional evidence for the validity of the "10^5 theory" in predicting infection follows:

- In 1955, Liedburg et al., demonstrated in rabbits that graft beds inoculated with $>10^5$ bacteria/ml destroyed skin grafts.[25]
- In 1956, Elek injected staphylococci into the skin of healthy human volunteers and determined that a mean of 7.5×10^6 staphylococci were needed to produce a pustule. The presence of a silk suture reduced the required bacterial load to 100 staphylococci.[10]
- Studies reported in 1964 and 1965 from the US Army Surgical Research Unit demonstrated and corroborated that burn wound sepsis required $> 10^5$ organisms/gm tissue.[26,38]
- Bendy et al. found that pressure sores did not heal until the microbial count was $< 10^6$ bacteria/ml.[4] According to Heggers and Robson, the discoveries of Bendy et al. and others led to the use of the chronic granulating wound as an experimental model to test the efficacy of various topical antimicrobial creams.[4,19,30]
- In 1957, Kass correlated the level of bacteruria with the presence or absence of symptomatic urinary tract infections. In 2000 patients with pyelonephritis, 95% had $> 10^5$ bacteria/ml of urine; 100% of symptomatic patients were found to have $> 10^5$ bacteria/ml of urine.[21]

- In 90% of the cases, positive cultures obtained by percutaneous transhepatic cholangiography from obstructed biliary tracts were derived from bile containing $> 10^5$ bacteria/ml.[13]

Additional literature to support the use of the "10^5 theory" in which this level of colonization has been correlated with the prognosis of skin grafts and surgical wounds follows:

- Krizek, Robson, and Kho evaluated 50 granulating wounds for skin grafting. The wounds were caused by venous stasis ulcers, trauma, or infection. Various topical agents were used to decrease bacterial counts. Biopsies were performed on alternate days. Those patients grafted when the wound biopsy showed $\leq 10^5$ bacteria/gm of tissue had a 94% graft take; those with higher counts had < 20% graft survival.[23]
- In another study, patients with pressure sores who were not candidates for standard flap closure were evaluated. All were treated with topical therapy, pressure relieving devices, and split thickness skin grafting if closure did not occur. Wound surveillance was performed frequently with quantitative bacteriology. [Technique entailed the following: (1) The wound surface was cleansed with isopropyl alcohol. (2) A biopsy of the open granulating wound was taken under sterile conditions with a dermal punch. (3) The tissue was weighed and homogenized after being diluted 1:10 with thioglycollate. (4) Serial dilutions were cultured on selective agar under both aerobic and anaerobic conditions.] In 14 cases, spontaneous healing occurred, but only after bacterial counts were sustained at 10^5 or fewer bacteria/gm of tissue. The remaining 18 were skin grafted, with a 92% graft survival in wounds with $\leq 10^5$ /gm of tissue and a > 80% graft loss in those with higher rates of bacterial colonization.[32]
- In a series of 41 cases in which surgical wounds had delayed primary closure or incisional abscesses that required drainage, treatment, and delayed closure, closure was successful in all cases with $< 10^5$ bacteria/gm of tissue.[32]

III. Discussion

The DAIDP reviewers found no evidence in the literature to clinically validate (that is, correlate with incidence and/or reduction of SSI) the surrogate microbiologic endpoints specified in the TFM for the patient preoperative skin preparation indication. Limitations of the literature review include the following. The literature search was limited to articles in the National Library of Congress' PubMed system and pertinent references derived from the articles' bibliographies. Search criteria included only the following key words: "skin," "surgery," "infection," AND "prevention." The reviewers were only able to thoroughly review articles written in English.

However, it should be noted that the reviewers screened greater than 400 articles and found only six that adequately met the pre-defined criteria of: (1) bacterial log reduction data available pre-scrub and at minimum post-scrub and (2) definition of SSI and rate of SSI during follow-up.

Though it does not provide direct evidence that the use of a patient preoperative skin preparation decreases the incidence of SSIs, the literature does provide supportive evidence for the role of quantitative bacteriology in other closely related settings and suggests that it may be possible to determine a minimal colony count on the skin at which SSIs are unlikely to occur. As the Agency moves forward in attempting to identify such a minimal level, several caveats should be kept in mind:

1. The literature suggests that contaminated foreign material entering the surgical wound, such as suture and drains, may decrease host resistance and promote infection at bacterial levels lower that 10^5.[10,34]
2. Robson's experience that beta-hemolytic streptococci may cause infection at a much lower innoculum than 10^5, possibly as low as 10^2.[33]

IV. Conclusions and Recommendations

In sum, the DAIDP reviewers have found no evidence to suggest that specific reductions in microbial counts on the skin, which are obtained with use of common preoperative skin preparation products, are associated with reductions in the incidence of SSI. The surrogate microbiologic efficacy endpoints required by the TFM for the patient preoperative skin preparation indication can not be considered to be validated as correlation with clinical outcome data does not exist. The literature review conducted by DAIDP staff confirms a similar finding made by the Hospital Infection Control Practices Advisory Committee (HICPAC) and published in the Guideline for Prevention of Surgical Site Infection in 1999.[27]

Options to move forward with identification and validation of surrogate microbiologic efficacy criteria for use in the TFM for the patient preoperative skin preparation endpoint include:

(1) Development of an animal model(s) in which a CFU threshold for relevant pathogens may be established below which SSI is unlikely to occur. While such data would not serve as clinical validation for use of the threshold in human studies, it would provide proof of concept for establishment of a threshold approach.
(2) Health outcomes research, that is, studies in human surgical patients in which the comparative efficacy of products could be demonstrated or in which a CFU threshold could be established at which SSI is unlikely to occur. A caveat when interpreting the results of these studies is that often the etiologies of SSIs are multifactorial.

In summary, additional research is needed to establish appropriate endpoints for studies in which topical products seeking the patient preoperative skin preparation indication are evaluated. This endeavor will require dedicated time and funds, and require the participation of both the FDA and FDA stakeholders.

Peter Kim, M.D.
Medical Officer
Division of Anti-Infective Drug Products

Jean Mulinde, M.D.
Medical Officer, Team Leader
Division of Anti-Infective Drug Products

Concurrence:

Janice Soreth, M.D.
Division Director
Division of Anti-Infective Drug Products

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