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Major Article

Observational study of sterile field bioburden levels during orthopedic arthroplasty surgery in operating rooms complying with current United States ventilation specifications

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Key Words:
Airborne microorganisms
Operating room air quality
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Periprosthetic joint infection
Risk factors

Background: Airborne contamination from microbe carrying particles (MCPs) is a risk factor for devastating early onset periprosthetic joint infection(PJIs). There are no published guidelines to quantify this risk factor for PJI events. This observational cohort project addresses this gap and utilizes a simple passive system to produce quantitative data from 80 total joint replacement cases performed in operating rooms built to current USA standards.

Methods: A petri dish-based system inspired by industrial cleanroom technology was deployed. Surgical helmet systems (SHSs) and strict protocols were used in all cases. 450 MCPs/m^2 was used as a cutoff for bioburden. This benchmark corresponds to the ultraclean air standard of 10 MCPs/m^3.

Results: 75/80 cases (94%) achieved desired benchmark levels of bioburden at the wound zone compared to only 52/80 (65%) of back table zones. No surgical site infections (SSI) or PJI events (0/80; 95% CI, 0.00−3.68%) at minimum 18-month (average 25.8 months) follow-up were detected.

Discussion: The current USA ventilation design uses low velocity airflow and appears to achieve ultraclean air conditions at the surgical site but requires SHSs and strict protocols. Higher contamination levels seen in back tables are consistent with this design.

Conclusions: This settle plate system may be useful for early onset PJI event investigations and thus lower the incidence of these complications.

BACKGROUND

Periprosthetic joint infections (PJIs) of hip and knee replacements are a subset of surgical site infections (SSIs) that cannot be treated with antibiotics or local wound care and require repeat surgeries and prolonged intravenous antibiotics. PJIs have dire consequences for patients and healthcare systems alike, affecting approximately 43,000 patients in 2022, and a projected 66,000 patients in 2030. Annual treatment costs are currently estimated at over $1.23 billion and are projected to rise to $1.88 billion 2030.[1] Therefore, minimizing the incidence of PJIs is a valuable healthcare system stakeholder goal.

The causes of PJIs are complex and multifactorial. Early onset PJIs occur within 90 days after surgery and are thought most likely related to the replacement procedure itself. Current AORN guidelines[2] recommend "an interdisciplinary team that includes an infection preventionist evaluate sterile technique practices" after an early onset PJI event. Many sterile technique practices involve protecting sterile fields from airborne microbial contamination. A recent international conference found in a consensus vote that intraoperative airborne microbe-carrying particles (MCPs) during hip and knee total joint replacement surgeries (TJRs) are a risk factor for PJI.[3]

Minimal sterile field bioburden is the purpose of operating room (OR) ventilation systems and sterile practices that address airborne contamination. How can an a PJI event investigative team assess if bioburden was minimal for a given case? Contemporary literature across multiple disciplines[4–8] does not address monitoring, quantification, or benchmarks for assessing

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intraoperative airborne bioburden. This report will attempt to address this gap first by reviewing basic science related to airborne microbial contamination.

The predominant source of airborne MCPs in the OR are derived from the personnel in the room.[9] The human MCP origins are nasopharyngeal droplets and squames.[10] Squames (which likely far outnumber droplets) consist of microbe-harboring desiccated human skin particles that are produced continually by natural epithelial turnover. The average human sheds these at a rate of 1,000 MCPs per minute. The equivalent diameters of MCPs can be modeled to a log-normal distribution, with an average diameter of 12 μm, fewer than 1% having a diameter smaller than 1 μm, and fewer than 5% having a diameter larger than 50 μm. This average size is comparable to water droplets in fog. The smallest particles, with movement governed by aerodynamic forces, can remain suspended in air for long time periods. The largest particles, with movement governed by gravitational forces, quickly settle out of the air onto surfaces. Particles of intermediate size are governed by both aerodynamic and gravitational forces.

Ventilation systems remove the smallest airborne particles from the OR; other particles fall directly or impinge onto sterile fields, nonsterile surfaces, or the floor. The problem in the orthopedic OR is that MCPs can be transported via airflow and turbulence and can be deposited onto the critical sterile zones.[11]

Several recent studies have examined operating room (OR) ventilation designs[12–14] and effects at the surgical site or protected zone but these have not considered the current USA standard design specified by paragraph 7.4.1 of ASHRAE 170-2021.[8] It is important to understand this design because there is no national requirement for orthopedic ORs to adhere to this standard resulting in the potential for large variability in US OR as-built ventilation systems. The ASHRAE 170 specification only applies to new and remodel projects. Existing OR ventilation systems are not automatically upgraded to this standard. Furthermore state, regional, or local authorities can modify these specifications and alter their implementation during construction.

Recent engineering studies demonstrate that current USA standards results in a unique airflow pattern over sterile fields.[15–17] See Figure 1 The central jet is low velocity (0.127–0.178 m/s; 25-35 FPM) HEPA filtered (hence sterile) air. This jet has a curved vertical profile and is limited to the center of the room beneath the ceiling air supply diffusers and over the surgical table. Heat loads from personnel and equipment influence the size of the central sterile zone due to buoyancy effects. The central jet is bounded by a free shear mixing layer that can allow MCPs from nonsterile ambient room air to enter the protected zone and impinge on the sterile field. The ambient air outside the central jet has much lower velocity allowing turbulent wakes from movement of personnel, equipment, and doors to transport MCPs to non-protected zones such as sterile back tables. Wakes and other effects can also transport MCPs to the free shear mixing layer resulting in critical zone contamination.

The issue of critical zone contamination is not unique to surgery or healthcare facilities. Cleanroom technology[18] has developed methods to address this in other fields such as pharmaceutical manufacturing. Two general cleanroom basic science principles are useful to understand the focus of this study. First: there are 2 major classes of air particulate matter that can be measured: viable and nonviable. A subclass are biologic particles that create fluorescence when struck by laser light sources. However, presence of a biofluorescence signal is nonspecific to a viable organism; that is, biologic particles may include both viable organisms and nonviable fragments. Nonviable particles pose no direct infection risk; therefore, this study is limited to measuring only viable microbes on squame particles.

The second cleanroom principle is there are the 2 broad classes of viable particle air quality measurement techniques. Active methods require air to move via a pump through a sampling device and to impinge on culture media. Passive techniques do not require pumped air flow and measure particles that sediment onto culture media. As shown above, the current USA ventilation design results in very low air velocities outside the central sterile jet. Active sampling instruments can in theory create unexpected backflows contaminating sterile fields in low velocity areas. This study uses settle plates, a passive system, therefore minimizing possible inadvertent contamination. A detailed discussion of several other test methods[19] that could be utilized for testing is beyond the scope of this paper.

This study was initiated to quantify the microbiological performance of operating rooms conforming to the ASHRAE 170-2021 standard under actual clinical conditions during orthopedic surgery. The

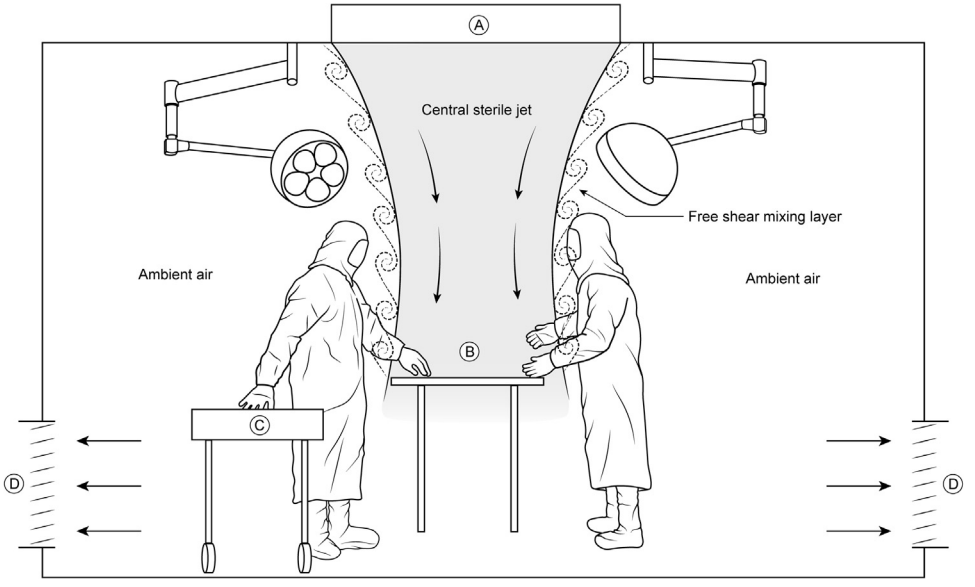

Fig 1. Schematic diagram of airflow in an operating room built to ASHRAE 170 – 2021 standards. (A) Ceiling diffusers supplying HEPA filtered air. (B) Surgical wound zone located in central jet of sterile air. (C) Back table zone for sterile instruments and supplies located in ambient air. (D) Near floor level exhaust grilles. Ambient air surrounds the central sterile jet and will contain variable amount of MCPs from personnel in room. The free shear mixing layer allows ambient room air to contaminate the central sterile jet.

clinical observation portion of the study follows STROBE guidelines and was performed for internal validation. This information may enhance the evaluation of SSI or PJI events at other facilities.

METHODS

The clinical study was accomplished via a prospective observational program approved by the institutional review board; informed consent was secured from all patients and surgeons. The tests were conducted from May 2016 through September 2017. Four surgeons performing primary total hip, knee, or knee revision cases at a single hospital participated. The surgeons used identical supplies and similar gowning and draping techniques. Surgical helmet systems (Stryker), AAMI Level 4 sterile gowns, surgeon indicator gloves, frequent regloving, impermeable drapes, minimal traffic, and jackets for non-scrubbed personnel were utilized in all cases. Except 1 revision case which used antibiotic loaded cement no additional antibiotics were used other than standard parenteral prophylaxis. Only normal saline was used for pulsatile irrigation.

During the normal course of their surgical practice, 5 cases were selected from each surgeon for testing spaced at 3 to 4-month intervals. The sample size of 5 cases was chosen to give an optimum balance of information and resource utilization. A total of 4 groups of 5 cases from each of the 4 surgeons were tested, for a total of 80 patients. The testing days were selected based on the surgeons' schedules, Petri dish availability, instrument logistics, and research staff schedule. The test schedule was blinded with respect to patient characteristics. The patients' intrinsic PJI risk factors of age, BMI, ASA class, and smoking status were recorded.

Two ORs were used for all cases. The OR construction documents were reviewed in detail to verify compliance with the current national codes. These rooms were nearly mirror images (approximately 57.0-m^2 floor area and 3.05-m ceiling height), with the Centers for Disease Control and Prevention (CDC)/ Healthcare Infection Control Practices Advisory Committee (HICPAC) -recommended airflow pattern (ceiling supply and exhaust near floor level).[7] The room air temperature, airflow rate, humidity, and pressurization relative to the entryway were monitored continuously and kept within regulatory requirements. Air was filtered by a rooftop unit with a pre-filter and high-efficiency particulate air (HEPA) filter and was ducted to 10 flat, perforated ceiling diffusers (61 cm × 122 cm, ASHRAE Group E non-aspirating type) arranged in a dispersed pattern See Figure 2.

The selected technique for MCP measurement utilized Petri dishes to capture MCPs that settled from the OR air, and is described in detail elsewhere.[20] This method examined 2 sterile fields: the back table zone, created for instruments and supplies, and the wound zone, created around the patient's operative extremity. Petri dishes were exposed from the time of sterile field creation until fascia closure. Plates were then collected and incubated for 48 h at 35°C, photographed, and the colonies were counted to determine the bioburden for each zone. Exposure time for each plate was recorded. Microbial deposition rate (MDR, MCPs/m^2/hour) and Microbial Deposition Total (MDT, MCPs/m^2) levels were calculated for each zone and case in a spreadsheet program. Since MDT allows easier comparison of case data (as opposed to reporting MDR and exposure time) only MDT values were used for subsequent analyses. The MDT benchmark for comparison is based on the historic orthopedic ultraclean air (UCA) limit of 10 MCPs/m^3.[21] This value corresponds roughly to a surface bioburden limit of 450 MCPs/m^2 (450 MDT).

Patient follow-up

At a minimum of 18 months post-surgery, each patient's medical record was reviewed for readmissions. The patients were also contacted by telephone to confirm the PJI status obtained from the medical records.

Statistical analyses

Patient and case descriptive statistics were calculated. Patient, surgeon, and case outcome variables were analyzed to establish patient risk factors and case MDT variation according to surgeon and test group in anticipation of PJI events. When assumptions of normality and homogeneity of variance were not met, variables were transformed with rank or Box-Cox transformation. MANOVA with follow-up univariate analysis was performed for significant findings. A paired t-test was performed on back table and wound zone MDT levels to confirm an expected difference seen in pilot studies. A Pearson's Product-Moment Correlation analysis was performed to assess the hypothesis of a relationship between bioburden and exposure time.

RESULTS

Patient descriptive statistical analysis for all 80 cases revealed an average age of 63.8 years (s.d. 9.04, range 42 − 79); ASA (American Society of Anesthesiology) class average was 2.58 (s.d. 0.49, range 2 − 3); and, the BMI (Body Mass Index, kg/m^2) average was 35.6 (s.d. 8.48, range 21 - 64).

All patients had medical record-based follow-up with average duration of 2.15 years (range, 1.56−3.76). Seventy-four out of 80 (92.5%) patients or families were available for telephonic follow-up. One patient expired during the study, 1.8 years after surgery, of unrelated causes. There were no readmissions for PJI or SSI (0/80; 95% CI, 0.00−3.68%). There were no patient safety events nor negative comments regarding the testing system itself.

Statistical analysis

Since there were no PJI or SSI related events there was no clinical significance in small variations between test and surgeon groups noted from the statistical analysis. However, it is described herein for completeness. The first multivariate analysis revealed statistically significant differences in patient characteristics across the surgeons and test groups. Follow-up univariate analysis showed there were significant differences in patient age ($F(3, 64)=3.07$, $P<0.05$, partial $\eta^2=0.12$) and patient BMI ($F(3, 64)=2.93$, $P<0.05$, partial $\eta^2=0.12$) among surgeon groups. However, there were no significant differences in patient ASA score ($F(3, 64)=0.21$, $P>0.05$, partial $\eta^2=0.10$) or patient smoking ($F(3, 64)=2.12$, $P>0.05$, partial $\eta^2=0.09$) among surgeon groups. There was a significant difference in ASA score between test groups ($F(3, 64)=2.95$, $P<0.05$, partial $\eta^2=0.12$). However, there were no significant differences in age ($F(3, 64)=1.58$, $P>0.05$, partial $\eta^2=0.06$), smoking ($F(3, 64)=1.45$, $P>0.05$, partial $\eta^2=0.06$), or BMI ($F(3, 64)=1.96$, $P>0.05$, partial $\eta^2=0.08$), between test groups.

A second multivariate analysis showed no statistically significant differences ($P>0.05$) in MDT values between surgeons or case groups. Because case groups were sequentially spaced at 3- to 4-month intervals, our study detected no statistically significant time "drift" variation.

The paired t-test showed a highly significant difference ($P<0.0001$) in MDT levels between wound and back table sterile zone groups. Regarding the back table sterile zone, Pearson's Product-Moment Correlation analysis detected no correlation between exposure time and MDT ($r=0.05$, $P>0.05$). As for the wound zone, exposure time was significantly and positively but weakly correlated with MDT ($r=0.41$, $P<0.01$).

Sterile zone bioburden data is detailed in Table 1. MDT outcomes for both sterile zones are plotted in the Figure 3 scattergram. Figure 4

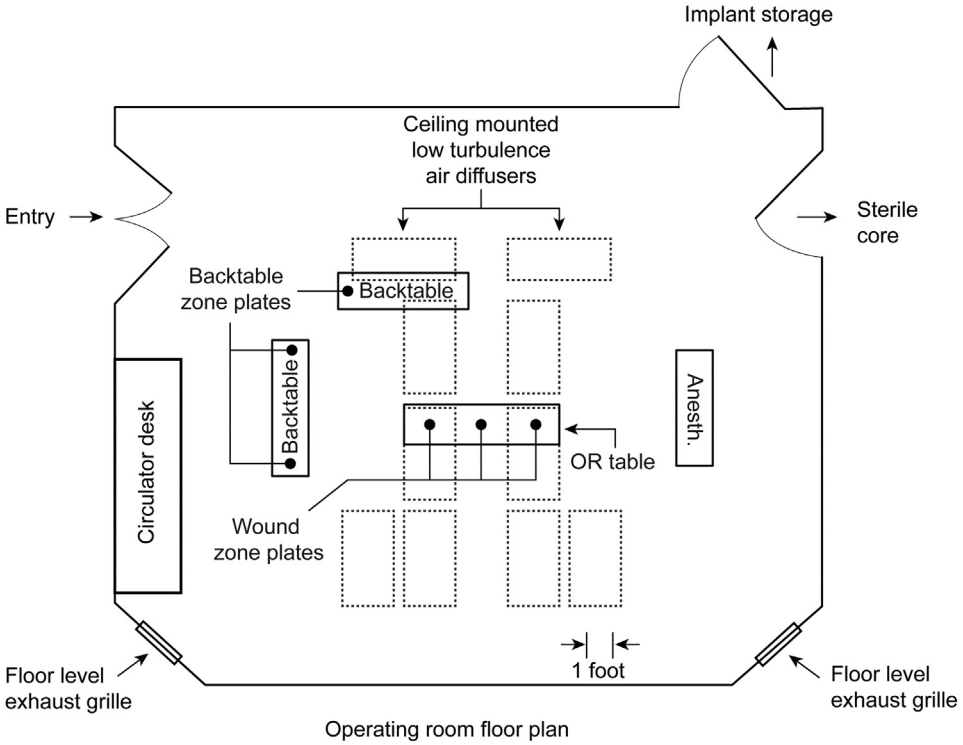

Fig 2. Plan view of operating room layout and plate positions generally used in study. Ceiling diffusers are shown as dotted rectangles. Only major equipment positions shown for simplicity. One OR in study had long axis of OR table aligned with long axis of diffuser array. This arrangement appears to meet the intent of ASHRAE 170 – 2021 paragraph 7.4.1b regarding diffuser coverage of surgical table. The second OR is shown and did not meet this requirement. No statistical differences in wound or backtable zones were noted between rooms.

focuses only on wound zone contamination data grouped by surgeon. Out of 80 cases, 75 (94%) had wound zone MDT under 450 colonies/m^2.

The 5 outlying cases with a wound zone MDT above this threshold were reviewed, and 4 cases involved patients with higher BMI (average= 43.2 kg/m2) who underwent direct anterior approach total hip arthroplasty. The remaining case was a primary knee replacement; review of settle plate photographs indicated one presumed fiber or hair fragment consistent with a possible surgical protocol violation. 52/80 (65%) cases had back table MDTs below 450. Fischer's Exact test showed that the difference in the 450 MDT standard complaince (wound zone, 94% versus backtable zone, 65%) was highly statistically significant ($P< 0.0001$).

DISCUSSION

The large variability in bioburden as shown in Figure 3 is a characteristic of microbiological systems. The sterile zone MDT values are consistent with ultra clean air (UCA) benchmark of 10 MCP/m^3 for 75/80 (94%) of cases. Overall, the ASHRAE 170-2021 ventilation system with low wall returns appears to provide UCA class protection to the sterile critical zone at the patient wound area. This study used multiple other procedures and special equipment including SHSs, AAMI Level 4 gowns and strict OR personnel protocols to achieve this protection level. The study design did not examine the effects of each component individually.

Table 1
Summary of sterile field bioburden and exposure times for all 80 cases.

Variable (units)	Minimum	Maximum	Mean	SD
Wound zone exposure (hours)	0.41	2.65	1.17	0.48
Backtable zone exposure (hours)	1.00	3.93	1.94	0.61
Wound zone MDT	0	942	205.1	165.57
Backtable zone MDT	0	1663	461.13	331.08

MDT, microbial deposition total (microbe carrying particles/m^2), SD, standard deviation.

The ASHRAE 170-2021 design with a diffuser face velocity of 25-35 fpm (0.127 -0.178 m/s) does not appear to meet unidirectional airflow (UDAF) ventilation system criteria proposed by Whyte[22] of 59-75 fpm (0.30-0.38 m/s). The ASHRAE 170-2021 design also does not appear to meet complex criteria for UDAF per DIN 1946[23] or ISO 14644-3:2019.[24] The ASHRAE 170-2021 design appears to meet the non-unidirectional (non-UDAF) pattern described by these same 3 references. Therefore, this report suggests that UCA class surgical site protection can be reliably obtained with a non-UDAF system and without the higher velocity airflows seen in UDAF systems (sometimes erroneously labeled "laminar" systems). This finding may be of interest to all ventilation designers and facility owners as lower velocities are associated with lower energy costs.

However, this energy savings requires a tradeoff in the size of the protective zone and protocol strictness. The protected zone is smaller than the supply diffuser ceiling area due to thermal effects that focus the sterile air in a smaller footprint at the OR table. The zone is bounded by a free shear mixing layer that was protected with the stringent procedures, AAMI Level 4 gowning, and SHS used in this study. Outside the protected zone is a much larger nonsterile zone of low velocity ambient air. This study shows that this zone, often containing back tables for instruments and supplies, has statistically significantly higher level of surface contamination (MDT > 450) above the UCA benchmark of 10 MCP/m^3. Only 65% of cases met the MDT benchmark for bioburden. Since the back table airborne wound contamination mechanism is indirect via transfer (as opposed to direct airborne inoculation in the wound zone) the clinical implications of this higher sterile field bioburden are concerning. Whyte[25] estimated 30% of wound contamination was due to the direct mechanism and

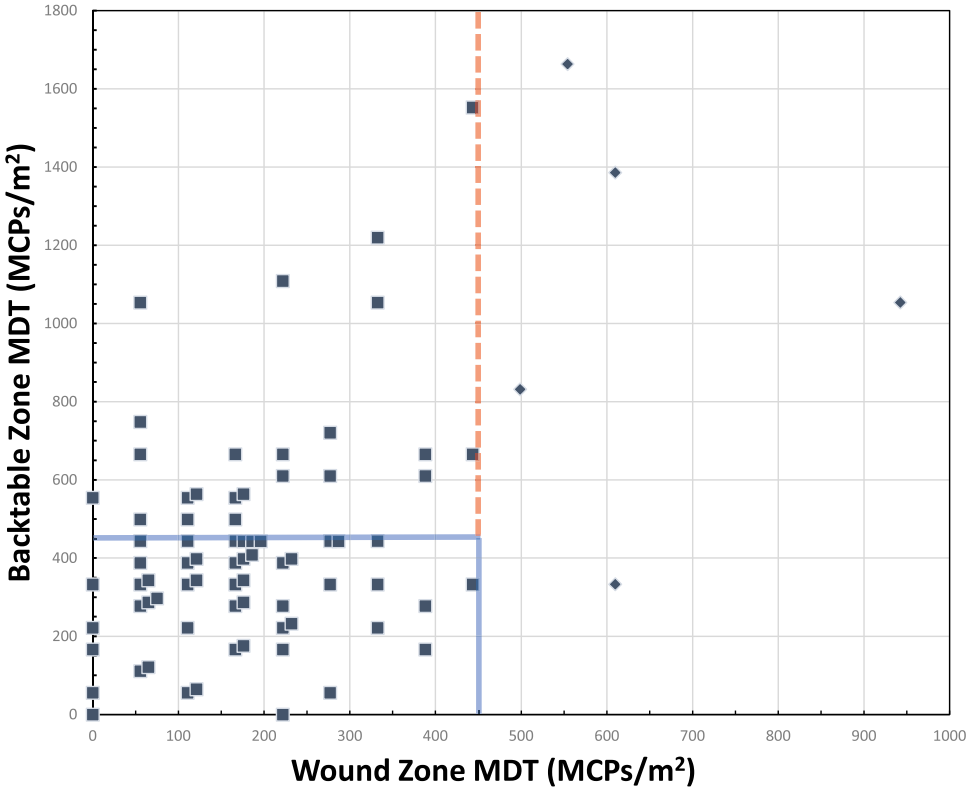

Fig 3. Each point represents one case. Wound zone MDT is plotted on the horizontal axis, and the back table zone MDT is plotted on the vertical axis. Overlapping data points have been offset slightly for clarity. The blue lines represent suggested MDT limits of 450 MCPs/m^2 which roughly correspond to Ultraclean air (UCA) limit of 10 MCPs/m^3. Points within 450 MCP limit in wound zone are shown as squares. Points to the right of the vertical dashed line had high wound zone MDTs exceeding 450 colonies/m^2 and are shown as diamonds. MDT, Microbial Deposition Total (MCPs/m^2).

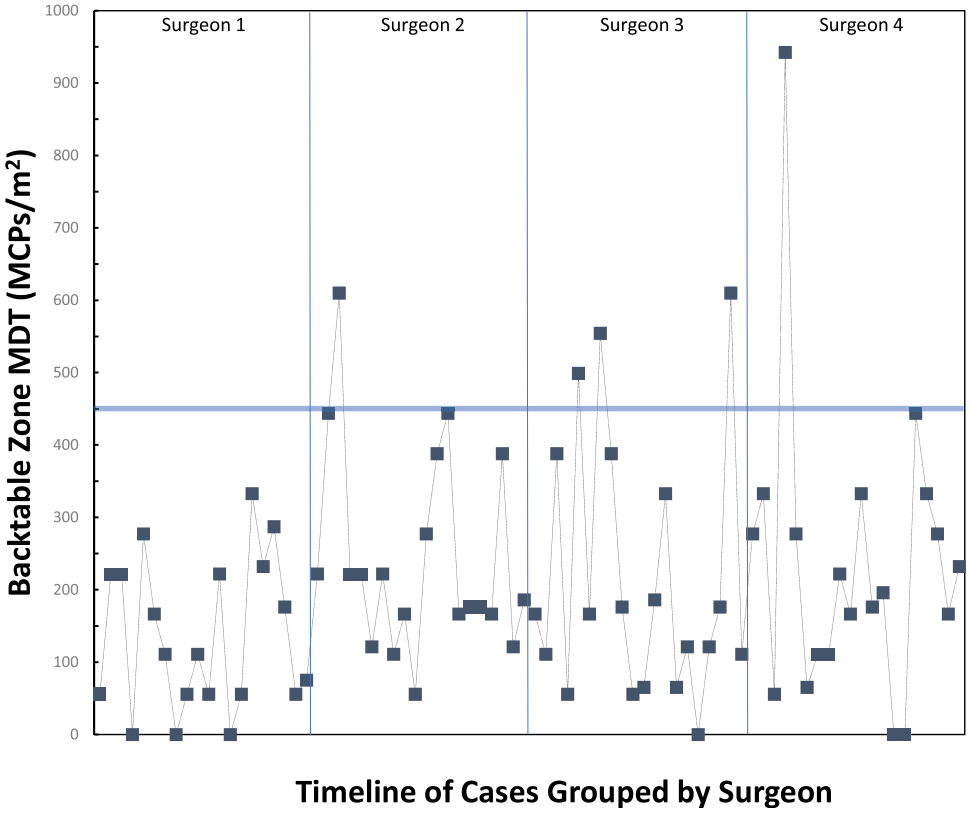

Fig 4. Each point on x-axis represents each of 20 cases performed by one surgeon. Cases are arranged in date and time order left to right. Y -axis is MDT level (MCPs/m^2). Blue line represents suggested MDT limit of 450 MCPs/m^2 which roughly correspond to ultraclean air (UCA) limit of 10 MCPs/m^3. This graph is similar to a classic Shewhart control chart.

thus implied 70% of wound bacterial contamination was from adjacent skin and the indirect transfer mechanism. Further study of back table transfer contamination risk mitigation appears indicated.

The technique and MDT data produce are consistent with a quality control sensor.[26] The data is produced within a few days of the index case, is likely proportional to the PJI risk, and measures viable particles in the direct causal chain of PJI events.

In addition, the outliers in the better controlled wound zone had explainable causes (case length, complexity, or protocol violation) for the higher MDT values. These characteristics suggest MDT data is suitable for a Shewhart type quality analysis chart. Figure 4 can be viewed as such a control chart that uses the UCA benchmark of 450 MDT as the control limit.

Microbiological testing as demonstrated in this study has several potential applications in PJI risk management and quality programs. If MDT levels appear stable, and outliers have explainable causes, then Figures 3 and 4 can be interpreted as acceptable benchmarks for the combined effects of OR ventilation and airborne sterile techniques. That is, most, but not all, MDT levels in the sterile zone meet the UCA/MDT standard of 10 MCPs/m^3 or 450 MDT. A PJI event occurring in an OR with similar performance is more likely related to other practices or factors than OR ventilation and sterile airborne contamination techniques.

The settle plate, data analysis, and test techniques used in this study were not created *de novo* but are based on widely accepted industrial cleanroom technology. Another USA healthcare example of applied cleanroom technology can be found in hospital compounding pharmacies.[27] The straightforward settle plate method utilized here has only become feasible recently due to advances in cleanroom technology in the form of terminally sterilized triple-wrapped Petri dish sets.[28] This method is both simple and inexpensive, with Petri dishes costing $4 each. Other costs include dish holder instrument sets, incubation facilities, and labor to read the plates and manage the testing program. However, a detailed economic analysis is beyond the scope of this report.

Study weaknesses

The uncertainty about the completeness of follow-up PJI data is not unique to this study. Despite having 100% chart follow-up, and despite additional telephonic confirmation, PJI events may have been missed due to patient relocation outside the facility's care area, reticence, or death with concurrent occult PJI during the follow-up period.

This was not a hypothesis-testing study, and so conclusions can be considered preliminary. The test method itself requires further development of standard operating procedures, performance checks, and data characterization (precision, accuracy, and detection limits) for quality control. The testing was conducted at a single institution, and a single investigator performed plate counts and compiled follow-up findings so confirmation bias could be present. Hence, the findings require replication at other sites for external validity.

However, this lack of higher-level evidence should not preclude technology innovations from being considered for reducing PJI incidence in patients and its impacts to the healthcare system[29] even in the absence of a well understood causal relationship. A recent Agency for Healthcare Research and Quality (AHRQ) report regarding improving patient safety states "Scarcity of evidence at a given point of time does not necessarily equal lack of effectiveness."[30]

CONCLUSION

The unique low energy consumption ventilation system specified for US operating rooms (ASHRAE 170-2021) produces different zones of sterile protection from airborne MCPs. This 80-patient observational study confirms a statistically significant difference in sterile field bioburden. Careful attention to equipment and techniques appears necessary to obtain UCA conditions (450 MDT, roughly 10 colonies/m^3) at the wound zone in 94% of cases. The back table zone in ambient low velocity air can be problematic with only 65% of cases meeting this standard. Surgical, quality, and infection control personnel may find this technique and benchmark information useful for future studies to improve patient outcomes.

References

1. Premkumar A, Kolin DA, Farley KX, et al. Projected economic burden of periprosthetic joint infection of the hip and knee in the United States. *J Arthroplasty*. 2020;36:1484–1489.e3.
2. Kyle E, ed. *Guidelines for Perioperative Practice*. Denver, CO: Association of periOperative Registered Nurses (AORN); 2022.
3. Aalirezaie A, Akkaya M, Barnes CL, et al. General assembly, prevention, operating room environment: proceedings of international consensus on orthopedic infections. *J Arthroplasty*. 2019;34:S105–S115.
4. Tubb CC, Polkowksi GG, Krause B. Diagnosis and prevention of periprosthetic joint infections. *J Am Acad Orthop Surg*. 2020;28:e340–e348.
5. Berríos-Torres SI, Umscheid CA, Bratzler DW, et al. Centers for disease control and prevention guideline for the prevention of surgical site infection, 2017. *JAMA Surg*. 2017;152:784–791.
6. The Association for Professionals in Infection Control and Epidemiology (APIC). *APIC Implementation Guide, Infection Preventionist's Guide to the OR [Internet]*. First edition Arlington, VA: The Association for Professionals in Infection Control and Epidemiology (APIC); 2015.. Accessed November 27, 2022.; https://jnjinstitute.com/sites/default/files/2018-12/093691-180619-APIC-Implementation-Prevention-Guide.pdf.
7. Guidelines for Environmental Infection Control in Health-Care Facilities. *Updated: July 2019. Recommendations of CDC and the Healthcare Infection Control Practices Advisory Committee (HICPAC). [Internet]*. Atlanta, GA: U.S. Department of Health and Human Services Centers for Disease Control and Prevention (CDC); 2003:30329.. p. 52,139. Accessed November 27, 2022.; https://www.cdc.gov/infectioncontrol/guidelines/environmental/index.html.
8. ASHRAE standing standard project committee 170, Cognizant TC: 9.6, healthcare facilities. ANSI/ASHRAE/ASHE standard 170-2021 ventilation of health care facilities. Peachtree corners, GA 30092; 2021.
9. Whyte W, Lytsy B. Ultraclean air systems and the claim that laminar airflow systems fail to prevent deep infections after total joint arthroplasty. *J Hosp Infect*. 2019;103:e9–15.
10. Popp W, Alefelder C, Bauer S, et al. Air quality in the operating room: surgical site infections, HVAC systems and discipline - position paper of the German Society of Hospital Hygiene (DGKH). *GMS Hyg Infect Control*. 2019;14:1–12.
11. Buchberg H. Improving the air environment of the operating room. In: *Proceedings of a Symposium on Clean Room Technology in Surgery Suites [Internet]*. NASA /Midwest Research Institute, Kansas City, Missouri; 1971.. Accessed November 27, 2022.; https://ntrs.nasa.gov/api/citations/19720020384/downloads/19720020384.pdf.
12. Cao G, Nilssen AM, Cheng Z, Stenstad LI, Radtke A, Skogås JG. Laminar airflow and mixing ventilation: which is better for operating room airflow distribution near an orthopedic surgical patient? *Am J Infect Control*. 2019;47:737–743.
13. Aganovic A, Cao G, Fecer T, et al. Ventilation design conditions associated with airborne bacteria levels within the wound area during surgical procedures: a systematic review. *J Hosp Infect*. 2021;113:85–95.
14. Lans JLA, Mathijssen NMC, Bode A, van den Dobbelsteen JJ, van der Elst M, Luscuere PG. Operating room ventilation systems: recovery degree, cleanliness recovery rate and air change effectiveness in an ultra-clean area. *J Hosp Infect*. 2022;122:115–125.
15. Zhai J, Hertzberg J, Smith W, Quinn G. *ASHRAE Research Project Report 1397 RP: Experimental Investigation of Hospital Operating Room (OR) Air Distribution*. Peachtree Corners, GA: ASHRAE; 2013:158. Report No.: 1397.
16. McNeill J, Hertzberg J, Zhai Z. Experimental Investigation of operating room air distribution in a full-scale laboratory chamber using particle image velocimetry and flow visualization. *J Flow Control Meas Vis*. 2013;1:24–32.
17. McNeill Jr. JS. *Experimental and Computational Investigation of Surgical Environment Air Distribution [Doctor of Philosophy]*. Boulder, CO: University of Colorado; 2020.
18. Whyte W. *Cleanroom Technology: Fundamentals of Design, Testing and Operation*. 2nd ed. Chichester, West Sussex, United Kingdom: John Wiley & Sons; 2010.
19. Working Group 023 of the IEST (The Institute of Environmental Sciences and Technology) Contamination Control Division (WG-CC023). IEST-RP-CC023: Microorganisms in Cleanrooms [Internet]. Schaumburg, IL: The Institute of Environmental Sciences and Technology (IEST); 2006. Accessed November 27, 2022. https://www.iest.org/Standards-RPs/Recommended-Practices/IEST-RP-CC023.
20. Harp JH. A clinical test to measure airborne microbial contamination on the sterile field during total joint replacement: Method, reference values, and pilot study. *JB JS Open Access*. 2018;3:e0001.
21. Lidwell OM, Lowbury EJ, Whyte W, Blowers R, Stanley SJ, Lowe D. Effect of ultraclean air in operating rooms on deep sepsis in the joint after total hip or knee replacement: a randomised study. *Br Med J Clin Res Ed*. 1982;285:10–14.

22. Whyte W, Mackintosh CA, Whyte WM. The design and testing of unidirectional airflow operating theatres. *Int J Vent*. 2021;0:1–16.
23. Working Committee NA 041-02-53 AA of DIN-Normenausschuss Heiz- und Raumlufttechnik sowie deren Sicherheit (DIN Standards Committee Heating and Ventilation Technology and their Safety). Ventilation and air conditioning – Part 4: Ventilation in buildings and rooms of health care, English translation of DIN 1946-4:2018-09. DIN-Normenausschuss Heiz- und Raumlufttechnik sowie deren Sicherheit (NHRS) DIN-Sprachendienst; 2018 Sep p. 74.
24. ISO Technical Committee 209. ISO 14644-3 Cleanrooms and associated controlled environments — Part 3: Test methods. Geneva: ISO (the International Organization for Standardization); 2019.
25. Whyte W, Hodgson R, Tinkler J. The importance of airborne bacterial contamination of wounds. *J Hosp Infect*. 1982;3:123–135.
26. Juran JM, Defeo JA. *Juran's Quality Handbook: The Complete Guide to Performance Excellence*. 6th edition New York: McGraw-Hill Professional Pub; 2010:1113.
27. The United States Pharmacopeial Convention. *USP 797, Pharmaceutical Compounding-Sterile Preparations. In: The United States Pharmacopeia 35th Revision: The National Formulary*. 30th ed. Rockville, MD: The United States Pharmacopeial Convention; 2012.
28. Tryptic Soy Agar - ICR 30ml | 146001 [Internet]. Accessed November 27, 2022. https://www.emdmillipore.com/US/en/product/Tryptic-Soy-Agar-ICR-30ml,MDA_CHEM-146001.
29. Busscher HJ, Alt V, van der Mei HC, et al. A trans-Atlantic perspective on stagnation in clinical translation of antimicrobial strategies for the control of biomaterial-implant-associated infection. *ACS Biomater Sci Eng*. 2019;5:402–406.
30. Agency for Healthcare Research and Quality (AHRQ), Department of Health and Human Services (HHS). Strategies to Improve Patient Safety: Draft Report to Congress for Public Comment and Review by the National Academy of Medicine [Internet]. Rockville, MD; 2020. Accessed November 27, 2022. https://www.pso.ahrq.gov/sites/default/files/wysiwyg/strategies-to-improve-patient-safety_draft-report.pdf.