European Journal of Parenteral & Pharmaceutical Sciences 2012; **17**(3): ??

Surface and air cleanliness in operating theatre environments

Gun Wirtanen[1]*, Salme Nurmi[2], Tapio Kalliohaka[2], Inga Mattila[2], Kimmo Heinonen[2], Seppo Enbom[2], Satu Salo[1] and Hannu Salmela[2]

[1] *VTT Expert Services Ltd, Espoo, Finland*

[2] *VTT Technical Research Centre of Finland, Tampere, Finland*

In this paper, airborne particles and microbiological hygiene are reported from a hospital hygiene study. The surface hygiene was monitored on operating theatre equipment, instruments and other adjacent surfaces as well as protective clothing and shoes. The study was performed from June until December 2010 in four Finnish operating theatres. The information obtained will be used in the development of a comprehensive package for hygiene assessment in both operating theatres and other hospital environments.

Key words: Operating theatre hygiene, surface hygiene, hygiene survey, airborne particles, aerobic bacteria, yeast, mould, coliform, *E. coli, S. aureus*.

Introduction

The Finnish healthcare sector is today undergoing structural change. A care guarantee and implementation of a cost-effective system have been driving forces. Finnish HighTech Hospital is a framework that boosts hygiene management and infection control in healthcare by benchmarking and adapting best working practices used in the pharmaceutical, electronics and food industries. The aim is to develop comprehensive packages for managing high levels of hygiene. The project recognises and assesses potential risks in the hygiene chain and suggests tentative proposals for improvement. Boosting the hygiene level reduces the number and costs of hospital epidemics, and also improves personnel and patient safety and the overall economy of the healthcare sector.

In general, Finnish hospital environments include a variety of buildings and premises from different decades. There are also a growing number of private clinics and hospitals that offer occupational healthcare. The operating theatres studied here were built between 1980 and 2010, and included recently renovated premises from both public hospitals and the private sector.

Objectives

The objectives of the study were to evaluate airborne particle concentrations and surface hygiene in typical operating theatre environments in four Finnish hospitals.

***Corresponding author:** Gun Wirtanen, VTT Expert Services Ltd, P.O. Box 1001, FI-02044 VTT, Finland; email: gun.wirtanen@vtt.fi; tel: +358 20 722 5222; fax: +358 20 722 7071

Methods

The methods included observation of practices and measurements used in clean production areas, e.g. in the electronics, pharmaceutical and food processing industries.

- Microbiological hygiene of operating theatre equipment, instruments and adjacent surfaces, and protective clothing, and of working practices as well as bioburden particles in the air.
- Sterility of instrument baskets stored for periods exceeding the recommended storage period.
- Cleanliness of laundry clean operation textiles.
- Cleanliness of operating theatres (particle concentration, pressure differences, cleanliness classes).
- Video exposure monitoring (VEM) was undertaken to follow particle release from different nursing practices. The video picture can be combined with different measurement data in order that different working practices can be examined or used for training purposes.

The number of samples analysed is shown in **Table 1**.

Microbiological surface hygiene

The microbiological hygiene in the four operating theatres was studied on both patient contact and non-contact surfaces, i.e. in the operating theatre, on prepared operating tables, on instruments, on clean hospital textiles and on hospital working clothes at various times during the working shift, using appropriate 3M™ Petrifilms (3M Microbiology Products, St. Paul, MN, USA). Aerobic bacteria, fungi, coliforms, *Escherichia coli (E. coli)* and *Staphylococcus aureus (S. aureus)* were recovered using the following media:

Table 1. Number of microbiologically analysed samples in the four operating theatres from June to December 2010.

Sample	Hospital 1	Hospital 2	Hospital 3	Hospital 4	Total number of sampling points
Air	41	63	47	29	180
Surfaces in operating theatres	308	641	433	166	1548
Workwear	189	192	187	180	748
Laundry clean textiles	81	84	169	42	376
Instrument tables	50	72	60	141	323
Sample total	669	1052	896	558	3175

Petrifilm™ Aerobic Count Plate (Petrifilm AC; Ø 50mm), 3M™ Petrifilm™ Yeast & Mold Count Plate (Petrifilm YM; Ø 70mm), 3M™ Petrifilm™ *E. coli*/Coliform Count Plate (Petrifilm EC; Ø 51mm) and 3M™ Petrifilm™ Staph Express Count Plate (Petrifilm STX; Ø 60mm). Petrifilm AC was incubated at 25°C for 5 days as were Petrifilm YM plates. The yeasts were easily differentiated from moulds based on growth characteristics and colours formed on the YM plates. The Petrifilm EC provided confirmed results on both coliforms and *E.coli* after incubation for 48h at 37°C. The results were read according to instructions given by the manufacturer. The Petrifilm STX provided *S. aureus* results after incubation at 37°C for 2 days. Typical colonies and high background flora were observed on the Petrifilm STX plates and both the 3M™ Petrifilm™ Staph Express Disk and API-Staph (bioMérieux, France) systems were used to identify *S. aureus* from all suspected colonies.

Airborne bioburden particles

Both the Klotz impactor FH5 (Markus Klotz GmbH, Bad Liebenzell, Germany) and the MAS-100 impactor (Merck KGaA, Darmstadt, Germany) were used in measuring the bioburden of the air. A volume of 200L air per sampling site was impacted onto the agar plates with an airflow of 30L/min when using FH5 or 100L/min when using MAS-100 in all four operating theatres and surrounding areas. Moulds and yeasts were analysed using sampling on potato dextrose agar (PDA, Difco 0013; Difco, BD – Diagnostic Systems, Sparks, MD, USA). Chlortetracycline (0.01%) and chloramphenicol (0.01%) antibiotics were used to suppress the growth of bacteria on this agar. The total amount of mesophilic bacteria was determined on plate count agar (PCA, Difco 0479). In this growth medium, cycloheximide (0.05%) was added to suppress growth of fungi. The plates were all incubated at 25°C for 5 days.

Sterility of instruments in stored instrument baskets

Reagents, materials and equipment required were brought into the clean area (laminar flow), before the analysis started. The stored instrument baskets were opened aseptically in the laminar area and the instruments were transferred into heavy duty Stomacher bags (Seward, UK). Sterile peptone saline was poured into the instrument bags and the samples were treated twice in the ultrasound bath for 5 min. The peptone saline was filtered through a water sampling funnel and the filter was aseptically placed on the plate count agar plate. The plates were incubated at 25°C for 5 days. The air quality in the laminar area was analysed using the Klotz FH5 impactor sampling 500L air on PCA plates. The measurement was repeated seven times during the sterility testing. These plates were also incubated at 25°C for 5 days. The air quality was also studied using an instantaneous microbial dectection (IMD) system (BioVigilant Systems Inc., Tucson, AZ, USA).

Airborne cleanliness with regard to particles

The airborne cleanliness classes of operating theatres with regard to particles were verified at rest conditions according to the standard EN ISO 14644-1, and also the EU GMP Guide (Volume 4, Annex 1). The particle concentrations were measured by a particle counter (MetOne 3313) and the pressure differences between operating theatres and surrounding areas were measured by a micromanometer. The particle concentrations were also measured during operation. In these cases, particles were measured only at one point near the exhaust.

The optical particle counter Met One 3313 was connected to the VEM system and used to determine the particle release from different surgical fabrics, caused by different nursing practices. The counter was used in the continuous mode where the total particle count (all particles >0.3μm) was obtained every 5 seconds. The relative variation of particle concentration was obtained using this system.

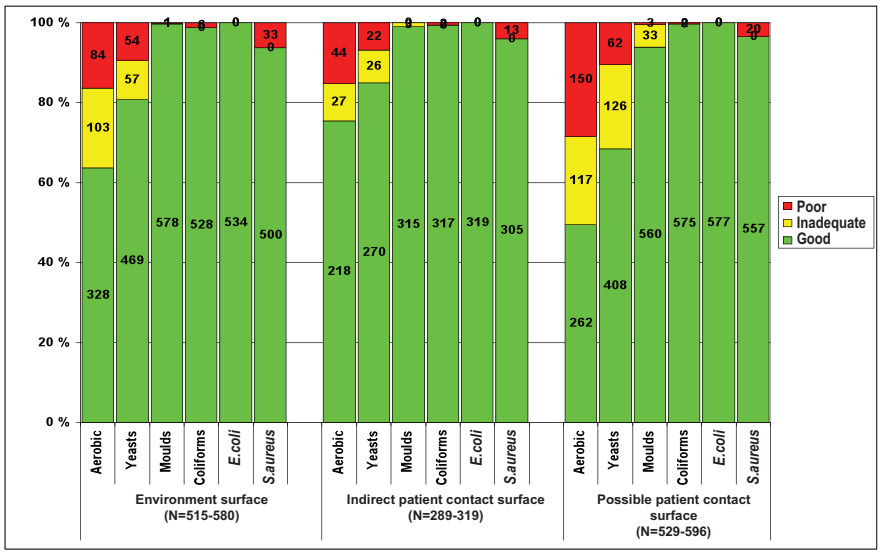
Results

Microbiological surface hygiene

Hygiene on operating theatre surfaces

The surfaces sampled in the operating theatres were divided into three groups: possible patient contact surfaces, indirect patient contact surfaces and environmental surfaces. Three levels for hygiene (good, inadequate and poor) were also set for all three groups. The higher the hygiene level, the fewer microbes were allowed on the surfaces. In **Figure 1** it can be seen that the surfaces were mostly contaminated with aerobic bacteria and yeasts.

Some surfaces had occasionally elevated numbers of bacteria on possible patient contact surfaces, e.g. support racks, sensors of electrocardiogram, tables for patient



Surface sample / Microbial level (cfu/Petrifilm)	Aerobic bacteria Good	Aerobic bacteria Inadequate	Aerobic bacteria Poor	Yeasts Good	Yeasts Inadequate	Yeasts Poor	Moulds Good	Moulds Inadequate	Moulds Poor
Environmental surface	< 20	20-50	> 50	< 10	10-25	> 25	< 10	10-25	> 25
Indirect patient contact surfaces	< 10	10-25	> 25	< 2	2-5	> 5	< 2	2-5	> 5
Possible patient contact surfaces	< 3	3-10	> 10	< 1	1-3	> 3	< 1	1-3	> 3

Surface sample / Microbial level (cfu/Petrifilm)	Coliforms Good	Coliforms Inadequate	Coliforms Poor	*E. coli* Good	*E. coli* Inadequate	*E. coli* Poor	*S.aureus* Good	*S.aureus* Inadequate	*S.aureus* Poor
Environmental surface	< 1	1-10	> 10	< 1	1-5	> 5	< 1	1-5	> 5
Indirect patient contact surfaces	< 1	1-5	> 5	< 1	1-2	> 2	< 1	1-2	> 2
Possible patient contact surfaces	< 1	1-2	> 2	< 1	1	> 1	< 1	1	> 1

Figure 1. The microbial levels of high hygiene surfaces in the operating theatres. These have been divided into three groups: possible patient contact surfaces, indirect patient contact surfaces and environmental surfaces. The microbial levels of each group have been defined as good, inadequate or poor depending on the microbial load and according to the levels given above.

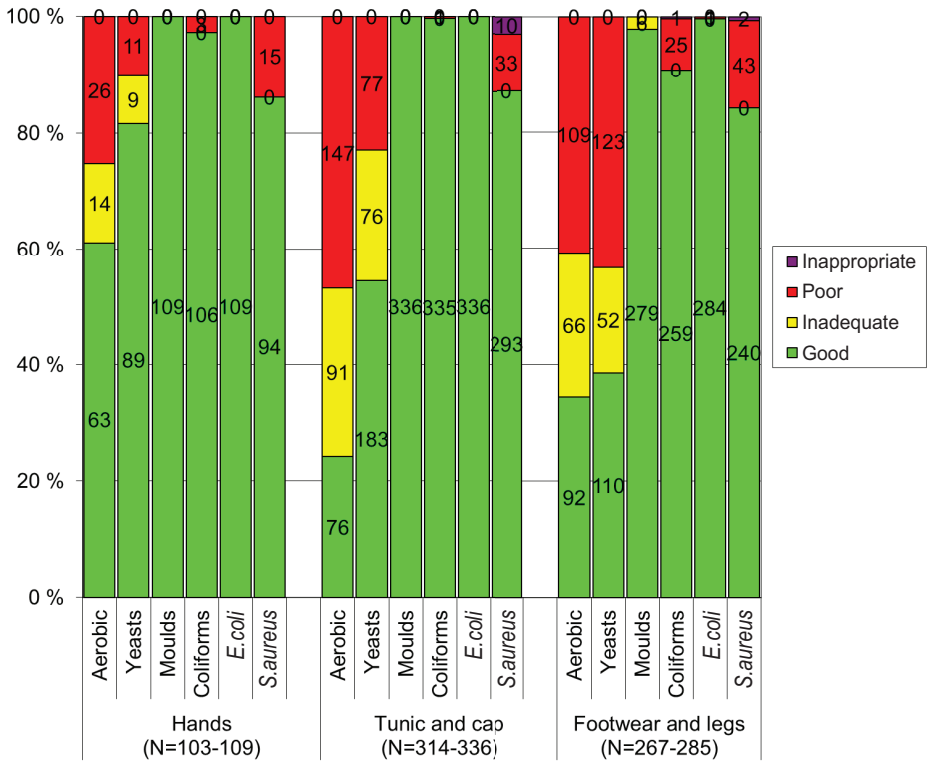
cleaning tools and anaesthesia equipment. Examples of indirect patient contact surfaces with occasionally elevated numbers of bacteria were computer keyboards, computer mice and stethoscopes. The following environmental surfaces had occasionally elevated numbers of bacteria: waste bin racks, floors, doors, cleaning tools, sinks and exhaust air ducts.

Workwear hygiene

The typical working suit in the four theatres consisted of V-neck tunic with short sleeves and trousers with ankle rib or stretch leg. This suit was made from a washable permanent-use textile and the material was microfibre woven fabric. The work footwear was versatile, including sandals or croc-type shoes. The suits were generally used all day, including outside the operating theatre department. Microbiological tests were undertaken on working clothes and shoes of three people 2–3 times during the working shift, i.e. at the beginning, the middle and/or at the end, during three working days. The sampling points were the upper front arm sleeve of the tunic, the pocket opening on the tunic front, the disposable cap, the front leg of the trousers, the upper surface of the shoe/sandal and the outer shoe/sandal sole. The hygiene levels (good, inadequate, poor and inappropriate) were set for all three workwear types. In the evaluation of workwear hygiene, one of the levels of indicator bacteria (coliforms and *E. coli*) and *S. aureus* was set as "inappropriate", which means that there were worryingly too many colonies on the surfaces studied. This rating was found especially on footwear surfaces and trouser legs but also on sleeves of tunics. The hygiene results of workwear are shown in **Figure 2**.

Aerobic microbes, yeasts, moulds, coliforms, *E. coli* and *S. aureus* were found on the footwear soles. During the working day, about 41% of the aerobic microbial count results were ranked poor, 26% inadequate and 32% good, clearly showing that footwear can transfer microbes from one place to another, from one department to another or from one hygiene zone to another. Operating theatre procedures should restrict the use of protective footwear to nominated areas. Strict procedures, including cleaning and disinfection practices, should be followed at all times.

The microbiological load was also quite high on tunics even at the beginning of shifts. During the working day, about 40% of the aerobic microbial count level was ranked poor, 30% inadequate and 30% good. Also, some yeasts and *S. aureus* were noticed on both tunics and caps. The microbiological level of cleanliness varied. Clearly, hygiene levels could be improved considerably, especially with respect to tunics and footwear, and regulations need to be sharpened.



Sampling places	Aerobic bacteria			Yeasts			Moulds		
Hygiene level (cfu/Petrifilm)	Good	Inadequate	Poor	Good	Inadequate	Poor	Good	Inadequate	Poor
Shoes and trouser legs	< 20	20-50	> 50	< 10	10-25	> 25	< 10	10-25	> 25
Tunic and caps	< 10	10-25	> 25	< 10	10-25	> 25	< 10	10-25	> 25
Hands	< 10	10-25	> 25	< 10	10-25	> 25	< 10	10-25	> 25

Sampling places	Coliforms			*E. coli*			*S.aureus*		
Hygiene level (cfu/Petrifilm)	Good	Poor	Inappropriate	Good	Poor	Inappropriate	Good	Poor	Inappropriate
Shoes and trouser legs	< 1	1-10	> 10	< 1	1-5	> 5	< 1	1-5	> 5
Tunic and caps	< 1	1-5	> 5	< 1	1-2	> 2	< 1	1-2	> 2
Hands	< 1	1-2	> 2	< 1	1	> 1	< 1	1-5	> 5

Figure 2. Hygiene of workwear used in the operating theatre. Workwear was divided into three groups: hands; tunic and caps; and shoes and trouser legs. The microbial levels of each group have been defined as good, inadequate, poor or inappropriate depending on the microbial load (see above).

Hygiene on instrument tables

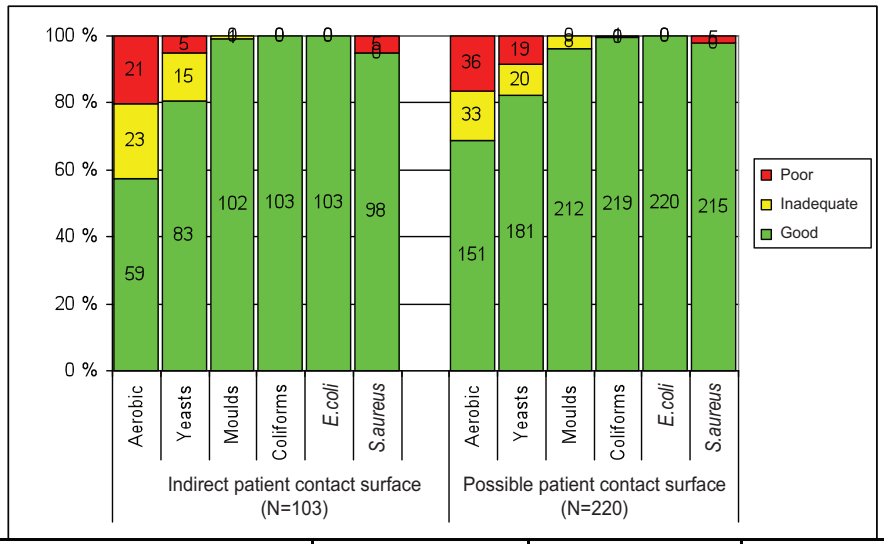
The instrument table surfaces sampled in the operating theatres were divided into two groups: indirect contact surfaces outside the laminar area and possible contact surfaces inside the laminar area. Here, the three hygiene levels were also set as good, inadequate and poor for both groups. The higher the hygiene level, the fewer microbes were allowed on the surfaces. **Figure 3** shows that the surfaces were clean. Any contamination found was mostly aerobic bacteria and yeasts. Occasionally elevated numbers of bacteria were found on instruments, table covers and surfaces of instrument tables. *S. aureus* was also found on instrument table covers.

Hygiene of clean laundry textiles and cleaning textiles

The microbiologically tested samples were analysed using three replicates of clean laundry textiles, such as operating bed cloth, patient lift cloths, bed linen, blankets, pillow cases, microfibre cleaning cloth for operating beds, microfibre cleaning cloths for floors, patient blankets, work tunics and work trousers. Large numbers of both aerobic bacteria and yeasts were recovered from cleaning cloths (results not shown). **Figure 4** shows that the microbes mostly found in laundry clean textiles were aerobic bacteria and yeasts. Contamination was occasionally found on all types of textiles. The hygiene level was sometimes classified as inappropriate where *S. aureus* was found on clean laundry textile surfaces (see **Figure 4**). In some cases, laundries did not use hoods on washed textiles transported to the hospitals, which could be a source of contamination before the textiles reach the hospital.

Airborne bioburden particles

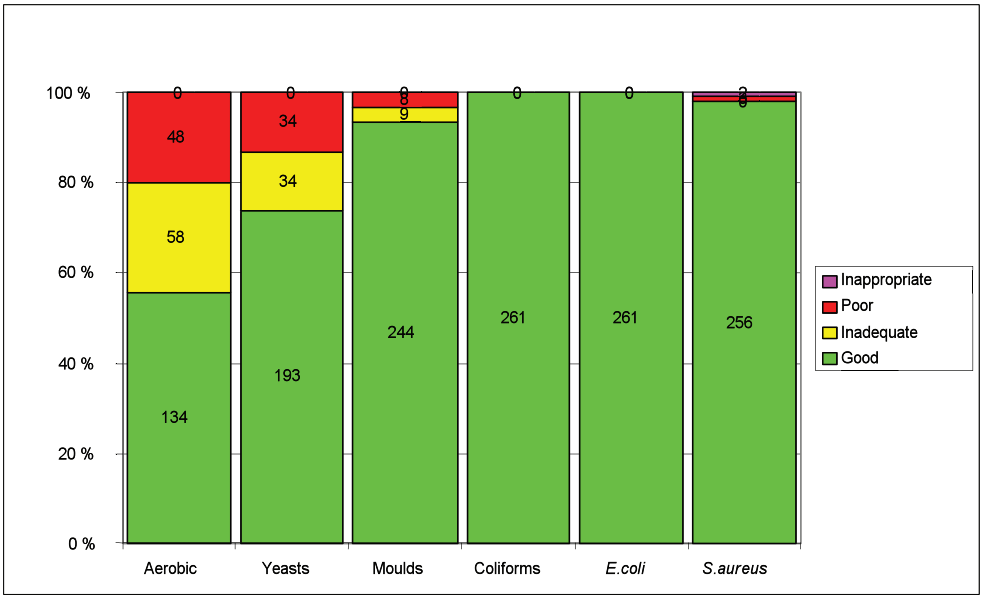
The air samples analysed (**Figure 5**) showed that >85% of sites evaluated had air of either good quality or slightly lower quality. Both the impaction and the optical particle counter methods showed similar trends in air quality. The levels for the high hygiene rooms in surrounding areas



Sampling points / Hygiene level (cfu/Petrifilm)	Aerobic bacteria Good	Aerobic bacteria Inadequate	Aerobic bacteria Poor	Yeasts Good	Yeasts Inadequate	Yeasts Poor	Moulds Good	Moulds Inadequate	Moulds Poor
Indirect contact surfaces outside the laminar area	< 2	2-5	> 5	< 2	2-5	> 5	< 2	2-5	> 5
Possible contact surface inside the laminar area	< 1	1	> 1	< 1	1	> 1	< 1	1	> 1

Sampling points / Hygiene level (cfu/Petrifilm)	Coliforms Good	Coliforms Inadequate	Coliforms Poor	*E. coli* Good	*E. coli* Inadequate	*E. coli* Poor	*S.aureus* Good	*S.aureus* Inadequate	*S.aureus* Poor
Indirect contact surfaces outside the laminar area	< 1	1-2	> 2	< 1	1-2	> 2	< 1	1-2	> 2
Possible contact surface inside the laminar area	< 1	1	> 1	< 1	1	> 1	< 1	1	> 1

Figure 3. The microbial levels of instrument table surfaces. The microbial levels of both groups have been defined as good, inadequate or poor depending on the microbial load on the surfaces sampled and according to the levels given above.



Microbe type/hygiene level (cfu/Petrifilm)	Good	Inadequate	Poor	Inappropriate
Aerobic bacteria	< 3	3-10	> 10	
Yeasts	< 1	1	> 1	
Moulds	< 1	1	> 1	
Coliforms	< 1		1	> 1
E. coli	< 1		1	> 1
S. aureus	< 1		1	> 1

Figure 4. The microbial levels of clean laundry textile samples (N=240–261) used in the operating theatre departments. These have been classified into four levels: good, inadequate, poor or inappropriate depending on the microbial load on the surfaces sampled and according to the levels given above.

were set based on the levels used in aseptic processing in the pharmaceutical industry.

Sterility of instruments in stored instrument baskets

The results from the stored instrument baskets showed that the contents of the baskets were sterile even after storage periods longer than now recommended in Finland (1 month). Contents of the baskets stored for both 7 and 9 months were all sterile. One of the sampled baskets stored for 5 months was contaminated, undoubtedly due to a small leakage in the plastic bag. Air sampling with both the Klotz impactor and the IMD equipment showed that no bacteria were recovered from air during the sampling period.

Airborne cleanliness of operating theatres

The results of the airborne cleanliness analysis are shown in **Table 2**. In some cases, the laminar ventilation system did not function properly and there were negative pressures periodically in the operating theatres. Negative pressure differentials can lead to contaminants moving from the environmental areas to areas in the high hygiene area. The cleanliness was classed as ISO class 6 in the case in which the pressure differentials were negative, but after changes in ventilation the air cleanliness improved to ISO class 5. The incorrect installations were thus immediately corrected.

The VEM analyses showed that the airborne particle concentration of particle diameter >1μm differed by 10–100 times in operating theatres during handling of different surgical fabrics and different nursing practices. It was not possible to determine any particular practices releasing short-term high concentration peaks in that particle size range.

The VEM analyses also showed that the use of diathermy equipment increased the concentration of particles 0.3–1μm in diameter remarkably (**Figure 6**). Short-term rises were up to several thousand times compared to the particle concentrations in the operating theatre at rest.

A snapshot of an airborne particle VEM analysis is shown in **Figure 6**. An example of short-term relative concentration variations in different particle size ranges can be seen.

Discussion

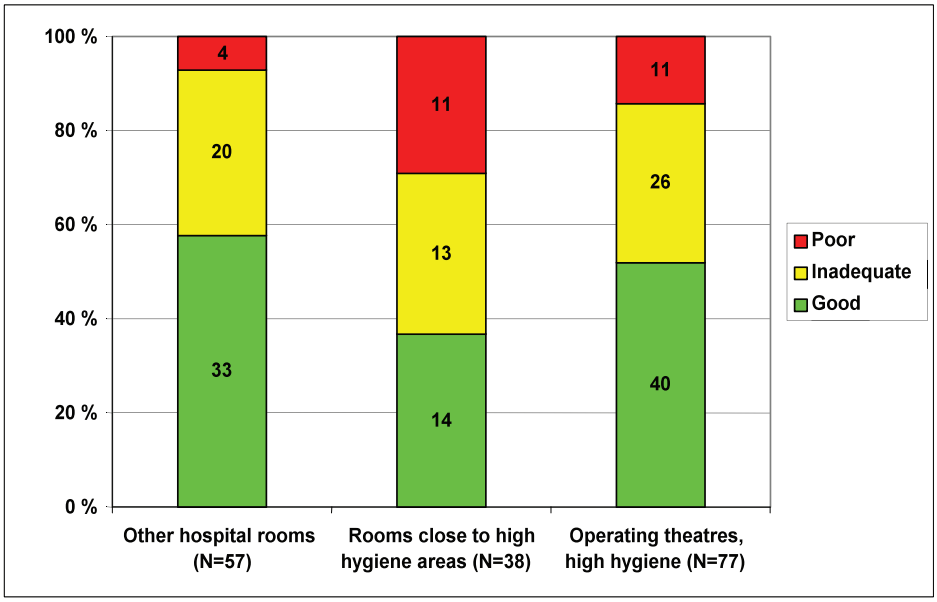
Microbiological surface hygiene

The results showed that footwear hygiene should be addressed in the operating theatre areas. Attention should be drawn to cleaning and disinfection practices of footwear in the operating theatre and usage should be restricted to nominated areas.

Operating theatre personnel clearly required additional training in hygiene matters; this should be reinforced at periodic intervals.

Airborne bioburden particles

Both the optical particle counter and the impaction method showed similar trends in the air quality, recording the size distribution of particles in the air within the operating theatre and its surrounding environment. The number of viable particles in the air should also be recorded.



Sampling place / Microbial level in air (cfu/200L)	Microbes: Good	Inadequate	Poor
Other rooms in hospital	< 5	5-20	> 20
Rooms close to high hygiene areas	< 2	2-5	> 5
Operating theatres, high hygiene areas	< 1	1-2	> 2

Figure 5. The airborne microbial levels. The evaluated air samples have been divided into three levels: high hygiene areas i.e. the operating theatres; the areas surrounding the theatres; and other hospital rooms. The microbial levels have been defined as good, inadequate or poor depending on the total microbial bioburden in the 200L sample taken.

Table 2. The results of the airborne particle cleanliness analysis.

Location/situation	ISO class	Pressure difference between operating theatre and corridor	Observation
Laminar area, at rest	5–6	-11–+43	Periodically negative pressure in operating theatres – ISO class 6
Area outside the laminar area, at rest	5–6	-11–+43	Periodically negative pressure in operating theatres – ISO class 6
Operating theatres with mixing ventilation, at rest	6	0–+5	
Operating theatres during operations	7–8	+3–+43	Emissions from operating personnel and operations
Surrounding areas	7–8		

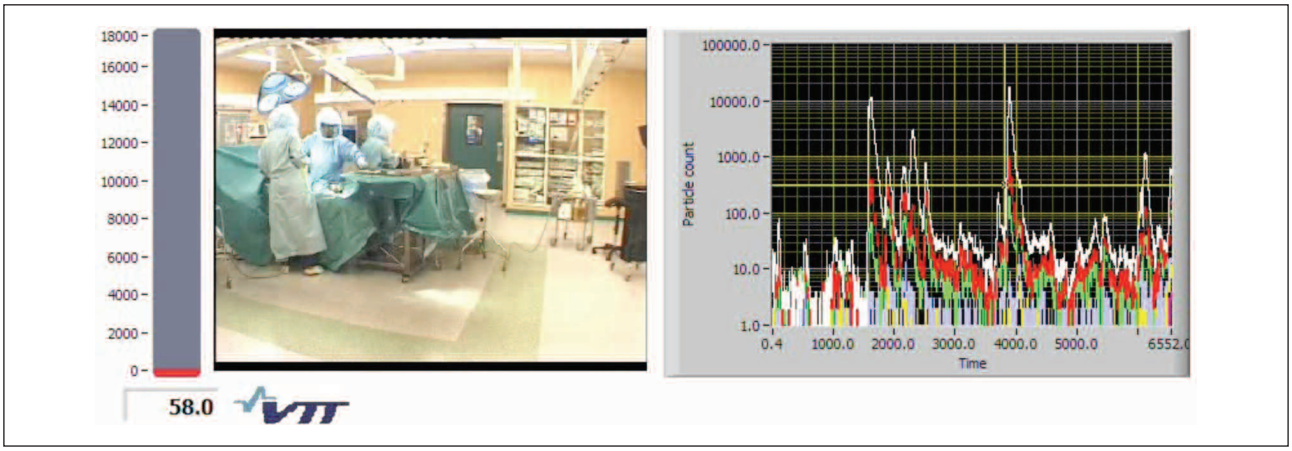


Figure 6. Variation of particle concentration during an orthopaedic operation.

Sterility of instruments in stored instrument baskets

Instruments in the stored baskets were found to be sterile after storage periods of 7 and 9 months[1]. Clearly, it is critical that the sterilised package is kept intact during storage. The tests showed that the hospitals could extend the storage time from the currently recommended 1 month to 3 or 6 months if required.

Airborne cleanliness with regard to particles

At rest conditions, the airborne particle cleanliness of operating theatres was normally ISO class 5 (laminar supply) or 6 (mixing ventilation), which conforms to the typical design values. In some cases, the air cleanliness with laminar supply was found to be ISO class 6 due to control problems in the ventilation systems. When these problems were resolved, the air cleanliness improved to class 5.

According to the VEM analysis, the particle concentration during operations was quite high. In some cases, the particle concentration was periodically even higher than in the surrounding corridor. This result indicates that it is very difficult to reduce the particle concentration in operating rooms by increasing ventilation over the typical rate of 20 to 40 air changes per hour.

Conclusion

The studies of airborne particles and microbiological surface hygiene were performed in operating theatres at four Finnish hospitals. The microbial contamination on different operating textiles seems to originate from personnel, additional microorganisms from other environmental surfaces, patients and laundering and other unspecified processes. The air particle measurements showed that the main sources of contamination are personnel again and the operations themselves. Therefore, the heating, ventilation and air-conditioning systems only have a limited effect in reducing particle concentration in these operating theatres whilst in use.

The information will be used in the development of a comprehensive package for hygiene assessment in operating theatres and other hospital environments[2].

Acknowledgements

The authors wish to thank the partners of the Finnish HighTech Hospital as well as other hospital partners contributing to the work. Microbiological analyses were carried out by VTT Expert Service Ltd in Espoo, Finland by Tuija Nieminen, Anneli Malmström, Susanne Nylander and Hannele Virtasalo. This work has been supported by Tekes, the Finnish Funding Agency for Technology and Innovation.

References

1 VTT Expert Services Ltd. VTT-S-04642-11. Research report on sterility testing of instruments in stored instrument baskets. Espoo, Finland: VTT Expert Services Ltd, 21 June 2011; p. 3 (in Finnish).

2. VTT Technical Research Centre of Finland. VTT-R-02058-12. Research report on hygiene management in high hygiene areas in hospitals. Tampere, Finland: VTT Technical Research Centre of Finland, 15 March 2012; pp. 58–66 (in Finnish).