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Comparison of droplet spread in standard and laminar flow operating theatres: SPRAY study group

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SUMMARY

Background: Reducing COVID-19 transmission relies on controlling droplet and aerosol spread. Fluorescein staining reveals microscopic droplets.
Aim: To compare the droplet spread in non-laminar and laminar air flow operating theatres.
Methods: A 'cough-generator' was fixed to a theatre trolley at 45°. Fluorescein-stained 'secretions' were projected on to a series of calibrated targets. These were photographed under UV light and 'source detection' software measured droplet splatter size and distance.
Findings: The smallest droplet detected was ~120 μm and the largest ~24,000 μm. An average of 25,862 spots was detected in the non-laminar theatre, compared with 11,430 in the laminar theatre (56% reduction). The laminar air flow mainly affected the smaller droplets (<1000 μm). The surface area covered with droplets was: 6% at 50 cm, 1% at 2 m, and 0.5% at 3 m in the non-laminar air flow; and 3%, 0.5%, and 0.2% in the laminar air flow, respectively.
Conclusion: Accurate mapping of droplet spread in clinical environments is possible using fluorescein staining and image analysis. The laminar air flow affected the smaller droplets but had limited effect on larger droplets in our 'aerosol-generating procedure' cough model. Our results indicate that the laminar air flow theatre requires similar post-surgery cleaning to the non-laminar, and staff should consider full personal protective equipment for medium- and high-risk patients.

Introduction

The coronavirus disease 2019 (COVID-19) pandemic has seen rapid developments in scientific and medical understanding of the SARS-CoV-2 virus [1—10]. Currently UK regulations are

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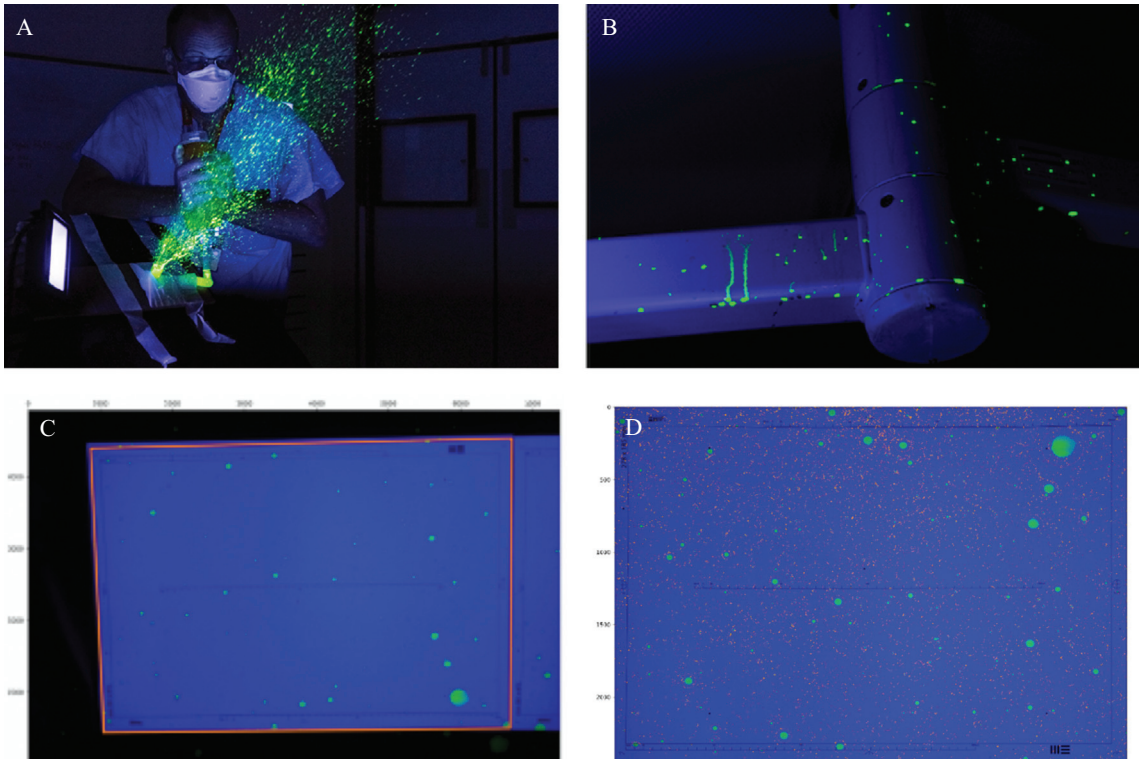

Figure 1. (A) The cough model. The nozzle was placed at 45° to the upright to mimic extubation; (B) showing the splatter on the operating lights of the laminar air flow theatre; (C) showing template mapping; (D) identification of spots by the Source Extractor Algorithm: the positions, brightness, and size of each spot were measured.

changing to keep pace with our scientific understanding, but there are gaps in the data, particularly around aerosol-generating procedures (AGPs) [1,2,8,11—13].

The UK National Health Service (NHS) and other healthcare systems face severe disruption, from efforts to protect both patients and staff from COVID-19 infection. Around 30—50% of capacity has been lost in the NHS due to these protective measures. We urgently need to understand the effect of AGPs on droplet and aerosol production within clinical environments in order to reduce disease transmission from patients with confirmed SARS-CoV-2 but also from patients and healthcare workers who may be asymptomatic carriers [14—17].

It is clear that SARS-CoV-2 may be spread by respiratory droplet splatter and subsequent hand/face contact, and that aerosols are also infective [9,14—17]. The accepted definition is that droplets have diameters >5 μm whereas aerosols have diameters <5 μm. Aerosols remain airborne for prolonged periods of time and can transmit the infection over large distances, whereas droplets fall rapidly to the ground [1]. However, this definition has come under increased scrutiny because particles >5 μm diameter may remain airborne for long periods of time and spread beyond 2 m [18—20].

Morawska *et al*. examined the size and distribution of droplets that are expelled from the respiratory tract [13]. For speaking and coughing, three modes in the aerosol and droplet size distribution were identified: two modes centred around 1—3 μm diameter and one mode centred around 100—200 μm diameter. Johnson *et al*. further developed this idea, finding three distinct peaks of droplets with diameters of 1.6, 1.7, and 123 μm during coughing [13]. They suggested that these peaks are associated processes: one in the lower respiratory tract, one in the larynx and upper respiratory tract.

Recently Brown *et al*. published a quantitative method of evaluation of aerosol generation during tracheal intubation and extubation [1]. They measured particles with diameters in the range 300 nm to 10 μm using a sampling funnel placed at 0.5 m away from the patient's face. Tracheal intubation produced very low quantities of aerosolized particles at 1.4 particles/L whereas extubation produced 21 particles/L. They compared these with a volitional cough, which produced 732 (SD: 418) particles/L. They made the point that intubation may not be an AGP at all and that the impact guidance around AGPs have increased waiting times for cancer and other surgeries [1].

However, larger droplets >200 μm are difficult to image and the particle analyser works best in very clean environments.

Simonds made a similar finding in patients undergoing non-invasive ventilation/nebulization and chest physiotherapy. Measuring droplets between 0.3 and 10 μm, they found that there was little aerosol generation and that most of the droplets fell to the ground within 1 m. However, in both these trials it was difficult to measure the trajectory of the larger droplets [21].

A standard operating room exchanges the air 20 times per hour and filters air with the removal of 80—97% of particles >5 μm. Laminar air flow systems equipped with high-efficiency particulate air (HEPA) filters remove 99.97% of particles >0.3 μm.

Current guidelines based on aerosol clearance times recommend a 20 min theatre clean for a non-laminar air flow theatre and a 2—6 min clean for a laminar air flow theatre.

Public Health England (PHE) guidance is that staff stand >2 m away from a high- or medium-risk patient.[12] However, the spread of larger droplets in such theatres has not been studied, and the importance of a deep clean between successive patients is unclear.

Fluorescent dyes have been used to mark body fluids, and to investigate the spread of infection [22—25]. Matava *et al.* developed a technique to assess the spread of droplets following extubation using a fluorescein dye [14]. They found that a clear plastic drape significantly reduces droplet/spray production from paediatric manikin.

There has been a research gap in the area of droplet research as there has not been a sensitive technique available to monitor aerosols or droplets from AGPs, and fomite spread once they have fallen on to surfaces, within clinical environments. This is of importance as a large percentage of healthcare workers infected in this way are asymptomatic and rapidly spread COVID-19 within clinical environments [26]. Our approach of using a fluorescein dye technique aims to fill this gap.

We developed a method of staining secretions with fluorescein, imaging with forensic photography, and analysing the images with a cosmological image-processing algorithm, usually used for detection of deep space objects such as stars and galaxies. With an extubation cough model, we compared the patterns produced by droplets falling on to paper targets, in operating theatres with laminar flow and standard ventilation systems.

Methods

A Laerdal manual resuscitator was used to blow air through a 17 cm corrugated catheter mount (internal diameter 15 mm). This was mounted on a theatre trolley ramped at 45° to simulate the typical position of a patient at extubation (Figure 1A) and placed at a height of 445 mm above and 445 mm to one side of the calibrated paper targets. A two-handed compression technique was used to mimic an extubation cough. The force of the cough was calibrated using a Peak Expiratory Flow Rate meter (Mini Wright Peak Flow Meter, Clement Clarke International, Harlow, UK).

A series of target sheets was aligned in front of the catheter mount, extending 3 m down the centre of the operating theatre, and under the canopy zone in the case of the laminar air flow theatre. The target sheets had calibrated scales printed on them to allow accurate image adjustment during analysis. A volume of 2.5 mL of 5% saline with a 1:20 dilution of 1% fluorescein minims (Bausch & Lomb, London, UK) was then injected into the catheter mount and simulated a cough, by compression of the Ambu bag. Once the splatter had occurred, the targets were imaged using a (Nikon DC 800) camera and an F80 lens. The camera was fitted with a UV flash and additional UV illumination was provided with two 30 W spotlights (Onforu, Guang Dong, China). Images were saved in numerical order and fresh plates were put out for each run of the experiment.

Some images of the cough simulation and of the theatre surrounds were also taken (Figure 1C, D). The test was repeated 11 times in non-laminar and laminar air flow theatres. We also calibrated the system using drops of a known volume between 0.1 and 2.0 μL. These were used to calculate the areas of splatter for a given drop size.

Observations were made within two operating theatres. The laminar air flow theatre has an ultraclean, vertical laminar flow ventilation system with HEPA filtration. The air under the canopy 'clean zone' is filtered and recirculated at an equivalent of 500—650 air changes per hour. It is discharged downwards resulting in an average air velocity of 0.38 m/s at 2 m above the floor and ≥0.2 m/s at 1 m above the floor. The cough source was under the canopy zone and the cough directed towards the centre of the zone. The second theatre meets requirements for a conventionally ventilated theatre (Department of Health guidance HTM 03-01 Part A). The air handling unit achieves 20—25 air changes per hour with supply air terminals at high level. Air temperature in theatres was set to 20°C and humidity between 40% and 60%.

Data analysis

The plates were positioned 445 mm below and 445 mm away from the cough source. An airborne particle is referred to as a 'droplet', and the region it covers on a detection plate as a 'spot'. The sequence of images from each test was uploaded to the Institute of Cosmology and Gravitation at the University of Portsmouth. The images were initially straightened and dewarped to correct for the position of the camera (Figure 1B). In these straightened images, one pixel has a width of ~85 μm, or an area of 7225 μm^2.

A source detection algorithm, Source Extractor, which is commonly used in astrophysics to identify objects in telescope images, was then used to detect individual droplet spots on the detection plates [27]. The algorithm was able to identify spots covering an area of ≥5 pixels, which corresponds to a spot of diameter 200 μm, or to droplet diameters of 120 μm. As well as identifying individual spots, the source detection algorithm also provides the basic properties of the spots, such as their size, position, shape, and orientation.

Statistical methods

All dot size measurements were tabulated by theatre type, cough, and distance. Total numbers of dots captured per cough, and total plate area covered per cough, from each type of theatre were compared, with null hypothesis of equal means. Our alternative hypothesis is that there are on average greater numbers of drops and coverage recorded on the plates in the non-laminar theatre. The standard deviations of spot counts and areas covered for each cough were of similar magnitude to the corresponding mean counts and areas. Therefore a randomized permutation test (non-parametric) was also performed under the null hypothesis of identical count and area distributions between the theatres, using the difference in means as the test statistic.

Tests were run using Statsmodels and NumPy (Python libraries). The spot size distribution was calculated by 'log-binning' spot area values (mm^2) from each theatre into a sequence of intervals of exponentially increasing width, and computing distance statistics (mean, variance, and standard error) for each bin. A similar method was used to generate a spot area vs distance plot for each theatre. Coverage statistics were computed for each plate distance and this was used to generate a distance—coverage plot for each theatre.

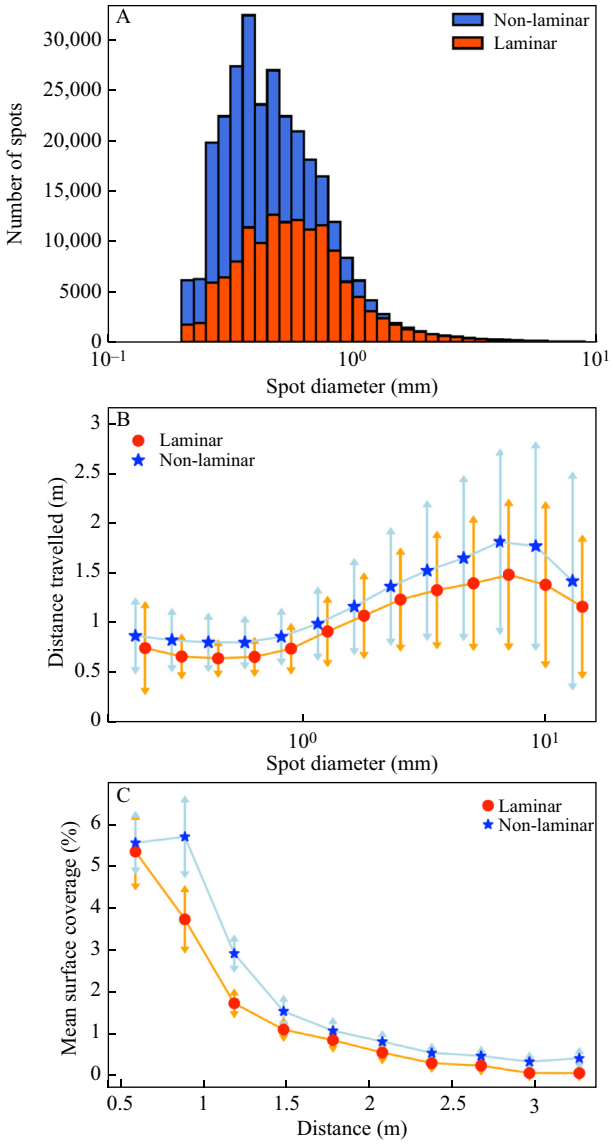

Figure 2. (A) Histogram of spot counts by diameter (mm) using diameter bin intervals ($10^{-1.5}$, $10^{-1.45}$, …, 10). The blue graph shows a large number of small drops in the standard theatre; the red graph, a reduction of smaller drops in the laminar theatre. (B) Distance travelled of drop vs diameter of spots. Points and bars show means and standard deviations of distance travelled by dots in each diameter bin. The further from the source the larger the average dot area became, indicating that the larger droplets had the momentum to travel further. In some plates large drops land and cause a splash, generating a range of small dots. (C) Surface coverage vs distance, showing that the laminar flow theatre displaced drops in every distance to 3 m, with significantly larger area covered by droplets in the standard theatre ($P < 0.02$). Error bars show standard errors in mean coverages.

Ethical review

This research was submitted to, and received support from, the University of Portsmouth Ethics Committee and the South Central — Berkshire NHS Research Ethics Committee. No patient data were collected during this research.

Results

The cough model was initially analysed to confirm that the cough peak flow accurately mimicked that of a normal human. A series of cough peak flows was measured: mean 351 L/min (SD: 22.7) with a range of 290–370 L/min.

The resolution of experimental splatter images was ~139.5 pixels per mm^2 and the smallest detectable spot was ~0.036 mm^2 in area, which, from the calibration graph, rendered a droplet diameter ~120 μm. The largest detected spot (475 mm^2) had an equivalent droplet diameter ~24,000 μm.

Whereas the counts of large spots are similar, the ventilation system of the laminar theatre displaced a fraction of the smaller droplets before they were able to reach the detection plates. These smaller droplets either deposited closer to the source or spread to other areas within the operating theatre. The mean numbers of spots per run were 11,430 (SD: 7882) in the laminar air flow theatre and 25,862 (SD: 8728) in the non-laminar ($P = 0.00016$).

The median spot diameter was 0.55 mm (laminar) and 0.45 mm (non-laminar). The full distribution of spot sizes is illustrated in Figure 2A as a histogram, binned by spot diameter, of the number of recorded spots per cough (averaged over all experimental repeats). From this it appears that the spot distribution differs between laminar and non-laminar air flows.

The mean distance of droplets resulting in small (diameter <1000 μm), medium (1000 μm < diameter < 2000 μm) and large (diameter >2000 μm) spots was measured. In the non-laminar air flow theatre, small droplets travelled on average 664 mm, medium 924 mm, and large 1282 mm; the laminar air flow theatre values were 814 mm, 1049 mm, and 1503 mm respectively ($P < 0.01$), indicating that there was a significant difference between the theatres at all droplet sizes. The maximum distance travelled in both theatres was >3.5 m.

Since the smaller droplets are most affected by the laminar flow ventilation, its effect on total area covered is less pronounced (Figure 2A). In the laminar air flow theatre the mean plate area covered was 8469 mm^2 (SD: 3775) and in the non-laminar air flow theatre was 11,818 mm^2 (SD: 3686). The corresponding *P*-value for the permutation test is $P = 0.022$.

There was a much slower decline in coverage at larger distances, where spots are typically several times larger than the median. The pattern may be understood by examining Figure 2B, which shows how the distance travelled by droplets depends on their corresponding spot diameter. The error bars in Figure 2B, which give the SD of the travel distance for each spot area, show that the range of smaller droplets (diameter <1 mm) is constrained to distances <1.5 m. The variation in the distances travelled by larger droplets is much larger, up to at least 3 m. Detailed information about travel distances is essential in order to build particle trajectory models that are consistent with realistic fomite splatter distributions.

The catalogue of spot areas and locations allows us to understand how the rate of fomite contamination varies with distance from the cough. Figure 2C shows how the mean plate fraction covered by spots varies with this distance. In both theatres there was a rapid decline in surface coverage. At 0.5 m the spot coverage was 5.55% non-laminar and 5.34% laminar ($P > 0.5$); at 1.2 m, 2.92% and 1.58%, respectively; at 2.1 m, 0.82% and 0.56%, respectively; and at 3.0 m, 0.34% and 0.08%, respectively.

Droplet splatter was also detected on the floor, walls, and operating theatre lights, and there was evidence of fomite transfer to light switches. The theatre lights were splattered in the laminar air flow theatres only. The lights in the laminar air flow theatres are positioned lower than in the non-laminar air flow theatres.

Discussion

Using a cough model, fluorescein 'body fluid' staining and image analysis can detect a wide variety of droplets both in terms of their size and velocity. On average 25,862 spots were detected in non-laminar and 11,430 spots in laminar air flow theatres, and a reduction of droplets in the laminar flow was identified. There was also a difference in the percentage of surface area affected, but this was less significant than the drop count, as the total area covered by larger droplets was similar in both theatres (Figure 2A).

Brown *et al*. investigated extubation in a laminar air flow theatre using an optical particle sizer that measured droplets with diameters from 0.3 to 10 μm, whereas we measured droplets with diameters from >120 μm with no upper limit [1]. They detected an average of 1310 smaller particles/L during a volitional cough. Perhaps a key difference was that they measured aerosol concentrations, whereas we measured deposited surface area. In our study, smaller particles were affected by laminar flow more than larger particles, consistent with their aerodynamic behaviour.

Direction is a key determinant of droplet distribution. Our cough model was directed upwards at a 45° angle, typical for extubation. In Brown *et al*.'s study, the patients were supine and the aerosols sampled at 50 cm from the patient. In the laminar air flow theatre the aerosols could have been affected by the air flow. Our data collection was limited to a strip of targets 210 mm wide extending directly in front of the cough model, and therefore we are unable to comment on droplets extending sideways from this.

Although these results were to some extent expected, it was surprising that large drops travelled further than smaller drops and could still travel >3 m within the laminar air flow theatre. By contrast with previously held views that large droplets fall rapidly to the ground, in our experiment many of the larger droplets had the momentum to travel >2 m [20]. It was also notable that larger droplets hit the ceiling and the surgical lamps in the laminar air flow theatre — though, due to the laminar air flow canopy, these were positioned lower as compared with the non-laminar. Within the laminar air flow theatre, the air is displaced sideways as it reaches the operating table, so there may have been more lateral dispersion of droplets and a wider target strip may capture more of the smaller droplets.

It seems that larger droplets are more resistant to the laminar flow and that guidelines for turnaround may need to be altered if the patient coughs during extubation. This demonstrates the importance of droplets in the spread of COVID-19 and focuses attention on the optimistic estimation from PHE that droplets fall to the ground within 1 m [12]. In our study, large droplets travelled up to 4 m, similar to McCool *et al*.'s study. These explosive coughs are perhaps best controlled with a physical barrier [28].

Our cough model does have limitations. For example, it does not measure aerosol production but the method could be used in conjunction with aerosol detection of AGPs, or in environments where accurate aerosol measurements are impossible. The respiratory tract has a more complicated configuration in comparison to our model, which only had a small (15 mm) external orifice. However, the droplet sizes produced and the distances they were projected were similar to human coughs. Larger droplets are mainly generated in the upper airway and the short, corrugated tube and 90° angle piece configuration of our model was effective at generating appropriate particle sizes. The key determinant of droplet projection is velocity, and our model reliably produced a clinically realistic cough peak flow of ~300 L/min [29]. Smaller droplets (diameter <120 μm) were not detectable with our technique due to the lack of spatial resolution of the initial imaging techniques. A further criticism is that we used saline 5%, which has a different viscosity (1.085 cP) to saliva (1.05 cP in women, 1.29 cP in men) [29]; however, Walker *et al*. have recently shown that the properties in forming aerosols and droplets are consistent and broadly similar [30].

Our results suggest that there is a reduction in droplet dispersion in laminar air flow theatres, but it is not clear whether this is enough to warrant preferentially undertaking AGPs in these theatres. To date, the most widely considered benefit of laminar flow theatres in the COVID-19 pandemic has been that 'downtime' after AGPs is minimized as aerosol clearance is comparatively rapid compared with conventionally ventilated theatres. However, more research is needed to understand the impact of laminar flow ventilation on droplets produced in a clinical situation, the use of physical barriers such as the AerosolShield (Birmingham, UK) or aerosol box on the spread of droplets and the importance of those droplets on the spread of SARS-CoV-2 [31]. The COVID-19 infection prevention and control guidance from PHE states that droplet precautions are 'measures used to prevent, and control infections spread over short distances (at least 1 metre or 3 feet) via droplets (greater than 5 μm) from the respiratory tract of one individual directly onto a mucosal surface or conjunctivae of another individual'; however, our data suggest that large droplets travel much further than this [12].

Previous studies have shown that better ventilation of spaces can reduce the airborne time of respiratory droplets [6]. There are uncertainties regarding the relative contributions of the different transmission pathways, but it is suggested that the engineering of indoor environments should target airborne transmission as one part of the strategy to limit infection risk indoors of viral infections such as COVID-19 [32].

In conclusion, we have developed a method of imaging droplets using fluorescein dye, forensic photography, and image analysis. Using a cough model, the spread of droplets through non-laminar and laminar air flow theatres has been investigated. Both theatres showed substantial droplet spread, beyond 2 m, during a cough simulation, but the distance travelled by the smaller droplets was reduced in the laminar flow theatre. These data may have an impact on current theatre protocols and could lead to the use of fluorescent dyes to stain all AGPs as an aid for hospital decontamination.

More research is urgently needed to map droplet spread within hospital environments. Most obviously this applies to areas where AGPs are performed but it is also important to understand that spread that will occur from uncontained coughing in all clinical settings. Combining droplet splatter analysis and optical particle sizing for smaller droplets and

aerosols will give us a better understanding of body fluid spread within hospitals. Furthermore, the possibility of creating a mathematical model of droplet spread within a three-dimensional map of each clinical environment may also be vital to predict droplet spread.

More research into AGPs is needed, as droplet spread could be wider than previously thought and current guidelines could be reviewed to reduce the potential of hospital infection from high-risk procedures.

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Conflict of interest statement
None declared.

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