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

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Key Words

Fluorescein,COVID-19, AGP, theatre, droplets, image analysis.

Abstract

Background

Reducing of COVID-19 transmission relies on controlling droplet and aerosol spread. Fluorescein staining reveals microscopic droplets. We used this technique to compare the droplet spread in a standard theatre (ST) and a laminar air flow theatre (LAF).

Methods

We used a 'cough-generator' fixed to a theatre trolley at 45-degrees. Fluorescein stained 'secretions' were projected onto a series of calibrated targets. These were photographed under UV light and a 'source detection' software measured droplet splatter size and distance.

Results

The smallest droplet detected was ≅ 120 μm and the largest ≅ 24,000 μm. We detected an average of 25,862 spots in the ST, compared with 11,430 in the LAF (54% reduction). The LAF mainly affected the smaller droplets (<1000 microns). The surface area covered with droplets was: 6% at 50 cm, 1% at 2m and 0.5% at 3 meters in the ST; and 3%, 0.5% and 0.2% in the LAF respectively.

Conclusion

Accurate mapping droplet spread in clinical environments is possible using fluorescein staining and image analysis. The laminar flow affected the smaller droplets but had limited effect on larger droplets in our AGP cough model. Our results indicate that LAF require similar post-surgery cleaning to those of ST and staff should distance 4m from 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 changing to keep pace with our scientific understanding [11,12] but there are gaps in the data [1,2,13] particularly around aerosol generating procedures (AGPs).[1,8]

The NHS and other healthcare systems face a year of 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 aerosol generating procedures on droplet and aerosol production with clinical environments to enable us to reduce disease transmission from both patients with confirmed SARS CoV-2 but also from patients and health care workers (HCWs) who may be asymptomatic carriers.[14-17]

It is clear that SARS-CoV-2 can be spread by respiratory droplet splatter and subsequent hand / face contact and aerosols are also infective.[9,14-18] 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,[1] whereas droplets fall rapidly to the ground. However, this definition has come under increased scrutiny because particles greater than 5 μm diameter can actually remain airborne for long periods of time and spread beyond two metres.[19-20] Instead a cut off of 100 μm has been suggested by Prather et al, as droplets with diameter > 100 μm tend to fall to the ground in seconds and follow ballistic trajectories.[21]

Morawska et al[13] explored the size and distribution of droplets that are expelled from the respiratory tract. 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[13] further developed this idea finding three distinct peaks of droplets with diameters of 1.6, 1.7 and 123 μm during coughing. 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[1] published a quantitative method of evaluation of aerosol generation during tracheal intubation and extubation. They measured particles with diameters in the range 300nm to

10 µm using a sampling funnel placed at 0.5m away from the patient's face. They found that tracheal intubation produced very low quantities of aerosolized particles at 1.4 particles L^{-1} whereas extubation produced 21 particles L^{-1}. They compared these with a volitional cough which produced 732 (SD 418) particles L^{-1}. 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 analyzer works best in very clean environments.

Simonds made a similar finding in patients undergoing non-invasive ventilation / nebulisation and chest physiotherapy. Measuring droplets between 0.3-10 µm they found that there was little aerosol generation and 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. [22]

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 HEPA (High-Efficiency Particulate Air) filters remove 99.97% of particles > 0.3 µm.

Current guidelines based on aerosol clearance times recommend a 20-minute theatre clean for a standard theatre and a 2-6 minute clean for a LAF theatre. Public Health England (PHE) guidance is that staff stand over >2m away from a high or medium risk patient.[12] However, the spread of larger droplets in such theatres have not been studied and the importance of a deep clean between patients is unclear.

Fluorescent dyes have been used to mark body fluids,[23-26] and investigate the spread of infection. Matava et al [14] developed a technique to assess the spread of droplets following extubation using a fluorescein dye. 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 onto surfaces, within clinical environments. This is of importance as a large percentage of HCW

harbour asymptomatic COVID-19. [27] 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 onto paper targets, in operating theatres with laminar flow and standard ventilation systems.

Methods

We used a Laerdal manual resuscitator to blow air through a 17 cm corrugated catheter mount (internal diameter 15 mm). We mounted this on a theatre trolley ramped at 45 degrees to simulate the typical position of a patient at extubation (Figure 1a). We placed this 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 were aligned in front of the catheter mount, extending three metres down the centre of the operating theatre, and under the canopy zone in the case of the LAF theatre. The target sheets had calibrated scales printed on them to allow accurate image adjustment during analysis. We then injected 2.5ml of saline with a 1:20 dilution of 1% fluorescein minims (Bausch & Lomb, London) into the catheter mount and simulated a cough, by compression of the Ambu bag. Once the splatter had occurred, we then imaged the targets 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 30w 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, 1d). The test was repeated 11 times in a standard theatre (ST) and a laminar flow theatre (LAF). We also calibrated the system using drops of a known volume between 0.1µl, and 2.0µl. These were used to calculate the areas of splatter for a given drop size.

Observations were made within two operating theatres. The laminar airflow theatre has an ultraclean, vertical laminar flow ventilation system with high efficiency particulate air (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 $m.s^{-1}$ at 2 m above the floor and not less than 0.2 $m.s^{-1}$ 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

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

We positioned the plates 445 mm below and 445 mm away from the cough source. We refer to an airborne particle as a 'droplet', and to the region it covers on a detection plate as a 'spot'. The sequence of images from each test were uploaded to the Institute of Cosmology and Gravitation at the University of Portsmouth. The images were initially straightened and de-warped to correct for the position of the camera (Figure 1b). In these straightened images, one pixel has a width of approximately 85 μm, or an area of 7.18 μm^2.

A source detection algorithm, Source Extractor,[28] which is commonly used in astrophysics to identify objects in telescope images, was then used to detect individual droplet spot on the detection plates. The algorithm was able to identify spots that were an area of 5 pixels or larger, 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

We tabulated all dot size measurements by theatre type, cough, and distance. We compared total numbers of dots captured per cough, and total plate area covered per cough, from each type of theatre, using Welch's t-test, 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, weakening the normality assumption underlying Welch's t-test. We therefore also carried out a randomized permutation test (non-

parametric) 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). We calculated the spot size distribution 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. We computed coverage statistics for each plate distance and used this to generate a distance-coverage plot, for each theatre.

Ethical review

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

Results

We initially analysed the cough model to confirm that the cough peak flow (CPF) accurately mimicked that of a normal human. We measured a series of CPFs, mean 351 l min^{-1} (SD 22.7) with a range of 290 to 370 l min^{-1}.

The resolution of experimental splatter images was approximately 139.5 pixels per mm^2 and the smallest detectable spot was approximately 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.

While 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 recorded per run were *11,430* (SD 7882) in the laminar theatre and *25,862* (SD 8728) in the non-laminar theatre. Our *t*-statistic, $t = 4.14$, for the difference of means gave a *p*-value $p = 0.00024$. The corresponding *p*-value for the permutation test was $p = 0.00016$.

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

We measured the mean distance of droplets which result in small (diameter < 1000 μm), medium (1000 μm < diameter < 2000 μm) and large (diameter > 2000 μm) spots. In the standard theatre, small droplets travelled on average 664 mm, medium 924 mm and large 1,282 mm, this contrasts to the laminar flow theatre which was 814 mm, 1,049 mm and 1,503 mm respectively. Welch's t-test with null hypothesis of equal means for each droplet size group in both theatres yielded *p*-values $p < 10^{-50}$ indicating that there was a statistical difference between the theatres at all droplet sizes. The maximum distance travelled in both theatres was over 3.5m.

Since it is the smaller droplets that are most affected by the laminar flow ventilation, its effect on total area covered is less pronounced (Figure 2a). In the laminar theatre we find that the mean plate area covered is $A_{lam} = 8{,}469$ mm^2 (SD 3775) and in the non-laminar theatre $A_{std} = 11{,}818$ mm^2 (SD 3686).

The corresponding *t*-statistic is $t = 2.15$; $p = 0.022$. The corresponding p value for the permutation test is also $p = 0.022$. These *p*-values, although larger, still provide strong evidence that the mean coverage of the detection plates was smaller in the laminar theatre.

We observed 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 standard deviation 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 3m. Detailed information about travel distances is essential in order to build particle trajectory models which are consistent with realistic fomite splatter distributions.

Our catalogue of spot areas and locations allows us to understand how the rate of fomite contamination varies with distance from the cough. In Figure 2c we show how the mean plate fraction covered by spots varies with this distance. In both theatres we observe a rapid decline in surface coverage. At 0.5m the spot coverage was 5.55% ST and 5.34% LAF (p>0.5), at 1.2m; 2.92% and 1.58% respectively, at 2.1m; 0.82% and 0.56% respectively and at 3.0m; 0.34% and 0.08% respectively.

We also detected droplet splatter on the floor, walls and operating theatre lights and evidence of fomite transfer to light switches. The theatre lights were splattered in the LAT theatres only. The lights in the LAT theatres are positioned lower than in the ST theatres.

Discussion

Using a cough model, we have demonstrated that our technique involving fluorescein 'body fluid' staining and image analysis can detect a wide variety of droplets both in terms of their size and velocity. On average we detected 25,862 spots in the standard theatre and 11,430 spots in the laminar flow theatre and were able to identify a reduction of droplets in the laminar flow theatre. There was a difference in the percentage of surface area affected as well 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[1] investigated extubation in a laminar flow theatre[1] using an optical particle sizer that measured droplets with diameters from 0.3 µm to 10 µm, whereas we measured droplets with diameters from greater than 120 µm with no upper limit. They detected an average of 1,310 smaller particles L^{-1} during a volitional cough. Perhaps a key difference was that they measured aerosol concentrations, whereas we measured deposited surface area. We know from our results that 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's paper the patients were supine and the aerosols sampled at 50 cm from the patient. In a LAF the aerosols could have been affected by the air flow. We limited our data collection to a strip of targets 210 mm wide extending directly in front of the cough model and therefore we are not able to comment on droplets extending sideways from this.

While these results were in some way expected, it was surprising that large drops could still travel over 3m within the laminar flow theatre. It was also notable that larger droplets also hit the ceiling and the surgical lamps in the laminar flow theatre although, due to the laminar air flow canopy, these were positioned lower as compared with the normal theatre. Within a laminar 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 does demonstrate the importance of droplets in the spread of COVID-19 and the optimistic estimation from PHE that droplets fall to the ground within 1m.[12] In our data large droplets could travel up to 4m, similar to McCool et al. These explosive coughs are perhaps best controlled with a physical barrier.[30]

Our cough model does have limitations. 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^{o} angle piece configuration of our model proved 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 around 300 litres per minute.[30] Smaller droplets (diameter < 120 μm) were not detectable with our technique due to the lack of spatial resolution of the initial imaging techniques. With greater resolution it will be possible to detect smaller droplets closer to the size of an aerosol. The calibration technique can be improved, assessing droplets with precise diameters. A further limitation is that the fluid we used was saline which has a lower viscosity than saliva and this could be resolved in further studies.

Our results suggest that there is a reduction in droplet dispersion in a laminar flow theatre, but it is not clear whether this is enough to warrant preferentially undertaking AGPs in laminar air flow theatres. To date the most widely considered benefit of laminar air flow theatres in the COVID-19 pandemic has been that 'downtime' after AGPs is minimised as aerosol clearance is comparatively rapid as compared to 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 Aerosol Box[31] on the spread of droplets and the importance of those droplets on the spread of SARS-CoV-2. The COVID-19 infection prevention and control guidance from the PHE state 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 5μm) from the respiratory tract of one individual directly onto a mucosal surface or conjunctivae of another individual"[12], however our data suggests that large droplets travel much further than this and a distance of 4m could be recommended.

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]

Conclusions

We developed a method of imaging droplets using fluorescein dye, forensic photography and image analysis. Using this with a cough model we have explored the spread of droplets through standard and laminar flow theatres. Both theatres showed significant droplet spread during a cough simulation, but the distance travelled by the smaller droplets was reduced in the laminar flow theatre. This 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 aerosol generating procedures are performed but it is also important to understand the spread that will occur from un-contained 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 3D mapped each clinical environment may also be vital to predict droplet spread.

Current guidelines could be adjusted to prevent contamination from high-risk patients, as our data suggests that a distance of 4m would be safer. More research in to AGPs is needed as droplet spread could be wider than previously thought and infection prevention and cleaning practices, especially in LAF theatres may need revision.

Authors Acknowledgements:

With thanks to Rosie Thomas, Dr Paul Smith, Dan McGuigan for photography; for software development, for Prof Chris Luca, Dr Michael Cottrell, Dr Jenny Child for editorial / methodology advice, Dr Andrew Lungren for software development.

Support:

This work was supported by an impact accelerator grant from the Science and Technologies facilities council. B.R.B. is supported by the Natural Environment Research Council (NE/P018459/1).

Competing interests:

The Authors declare no competing interests,

Financial Support Grants:

This work was supported by an Impact Accelerator Account; Science and Technology Facilities Council. AA is supported by the Royal Society Wolfson Fellowship.

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Figure 1

Figure 1a The cough Model in action, the nozzle was placed at 45 degrees to the upright to mimic extubation; **Figure 1b**, showing the splatter on the operating lights of the laminar air flow theatre; **Figure 1c** showing template mapping; **Figure 1d** identification of pixels by the Source Extractor Algorithm, the positions, brightness and size of each slat were measured.

Figure 2

Figure 2a Histogram of spot counts by diameter (mm) using diameter bin intervals $(10^{-1.5}, 10^{-1.45}, \ldots, 10)$. 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. **Figure 2b.** 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. **Figure 2c** Surface coverage vs distance, showing the laminar flow theatre displaced drops in every distance to 3m, with significantly larger area covered by droplets in the standard theatre ($p < 0.02$). Error bars show standard errors in mean coverages.