OPEN ACCESS

Original research

Aerosol emission from the respiratory tract: an analysis of aerosol generation from oxygen delivery systems

Fergus W Hamilton ,[1,2] Florence K A Gregson,[3] David T Arnold ,[4] Sadiyah Sheikh,[3] Kirsty Ward,[5] Jules Brown,[6] Ed Moran,[7] Carrie White,[8] Anna J Morley,[4] AERATOR Group, Bryan R Bzdek,[3] Jonathan P Reid,[3] Nicholas A Maskell,[4] James William Dodd [2,4]

► Additional supplemental material is published online only. To view, please visit the journal online (http://dx.doi.org/10.1136/thoraxjnl-2021-217577).

For numbered affiliations see end of article.

Correspondence to
Dr Fergus W Hamilton, MRC-IEY, University of Bristol, Bristol BS10 5NB, UK;
fergus.hamilton@bristol.ac.uk

FWH and FKAG contributed equally.
NAM and JWD contributed equally.

Received 4 May 2021
Accepted 17 October 2021
Published Online First
4 November 2021

ABSTRACT

Introduction continuous positive airway pressure (CPAP) and high-flow nasal oxygen (HFNO) provide enhanced oxygen delivery and respiratory support for patients with severe COVID-19. CPAP and HFNO are currently designated as aerosol-generating procedures despite limited high-quality experimental data. We aimed to characterise aerosol emission from HFNO and CPAP and compare with breathing, speaking and coughing.

Materials and methods Healthy volunteers were recruited to breathe, speak and cough in ultra-clean, laminar flow theatres followed by using CPAP and HFNO. Aerosol emission was measured using two discrete methodologies, simultaneously. Hospitalised patients with COVID-19 had cough recorded using the same methodology on the infectious diseases ward.

Results In healthy volunteers (n=25 subjects; 531 measures), CPAP (with exhalation port filter) produced less aerosol than breathing, speaking and coughing (even with large >50 L/min face mask leaks). Coughing was associated with the highest aerosol emissions of any recorded activity. HFNO was associated with aerosol emission, however, this was from the machine. Generated particles were small (<1 µm), passing from the machine through the patient and to the detector without coalescence with respiratory aerosol, thereby unlikely to carry viral particles. More aerosol was generated in cough from patients with COVID-19 (n=8) than volunteers.

Conclusions In healthy volunteers, standard non-humidified CPAP is associated with less aerosol emission than breathing, speaking or coughing. Aerosol emission from the respiratory tract does not appear to be increased by HFNO. Although direct comparisons are complex, cough appears to be the main aerosol-generating risk out of all measured activities.

Key messages

What is the key question?
► Do high-flow nasal oxygen (HFNO) and CPAP produce clinically relevant aerosols?

What is the bottom line?
► In healthy volunteers, CPAP produced no aerosol, and HFNO produced no clinically relevant aerosol, while coughing was associated with significant aerosol production.

Why read on?
► Management of patients in respiratory failure with potentially infectious pathogens remains a complex area with little evidence. This paper provides some of the first high-quality data on potential risks associated with aerosol emissions.

INTRODUCTION

The WHO describes disease transmission through three routes: physical contact, 'droplet' inhalation (larger particles which settle in a reasonably short distance) or 'airborne' (smaller particles which travel as aerosols on air currents, remaining in the air for longer and distributing over a wide area).[1] SARS-CoV-2, the virus that causes COVID-19, can be transmitted via aerosols with aerosol emission being the putative mode of transmission for many super-spreading events.[2 3] Although the exact size of aerosol particles responsible for airborne transmission (and the ability of virus to survive in these particles) continues to be debated, it is clear that the dispersion of particles smaller than 5 µm is largely determined by the room ventilation (air exchange) rate, thereby posing a potential risk to include those not in close contact, especially in poorly ventilated areas.[4] Quantifying the concentration of particles of this size range is therefore critical for understanding the risk of disease transmission.

Traditionally, medical procedures are deemed as 'aerosol generating' when there is a perceived risk of increased generation of aerosol from the patients' mucosa or respiratory tract compared with normal breathing, exposing staff and others in the vicinity to risk of inhalation of aerosolised airborne virus. For these aerosol-generating procedures (AGPs), an extra set of infection control precautions are mandated.[5–7] These additional precautions often involve: segregating these patients from others, changing personal protective equipment to include FFP3 (or N95) masks that limit aerosol inhalation rather than fluid-resistant surgical masks

Linked

► http://dx.doi.org/10.1136/thoraxjnl-2021-218035

Check for updates

To cite: Hamilton FW, Gregson FKA, Arnold DT, *et al*. *Thorax* 2022;**77**:276–282.

(FRSMs), ensuring adequate ventilation, and allowing 'fallow' time between procedures to allow aerosol to disperse.

These mitigation strategies have significant impact on healthcare capacity, costs and potential harms; it is therefore critical to accurately identify whether these procedures truly do generate aerosol.[8] Our aim was to identify whether procedures generate appreciable aerosol and whether the aerosol number concentration is lower than that generated by a cough. If so, the AGP is likely of low risk and misclassified.

Oxygen delivery and respiratory support including continuous positive airway pressure (CPAP) and high-flow nasal oxygen (HFNO) are used for the management of hypoxaemic respiratory failure complicating COVID-19 pneumonia. CPAP and HFNO are currently deemed AGPs by both the WHO and Public Health England (now the UK Health Security Agency), although the evidence for these recommendations is sparse.[5 7 9 10] Current guidance stipulates the need to cohort these patients and universal FFP3 usage in any setting where a patient is receiving CPAP or HFNO. However, universal FFP3 usage is not currently recommended when caring for general inpatients with COVID-19, despite the potential risks from breathing, speaking and coughing in this setting.

In this study, we set out to quantify aerosol generation in both CPAP and HFNO and compare it with breathing, speaking and coughing without these supports.

METHODS

Full technical methods are in the online supplemental appendix S1. In brief, healthy volunteers were recruited in ultra-clean, laminar flow operating theatres and underwent a protocolised set of testing (breathing, speaking and coughing) under different oxygen delivery systems and without respiratory support. Participants were instructed to perform three voluntary coughs into the measuring funnel, moving their head away from the funnel after each cough.

Aerosol measurements were taken simultaneously by two separate devices, the Aerodynamic Particle Sizer (APS) and Optical Particle Sizer (OPS) (both manufactured by TSI), via a 3D-printed funnel and through 0.45 m of conductive silicone tubing. Both devices were included as they work on differing technologies and are able to detect aerosols of differing sizes (APS, 0.5–20 µm; OPS, 300 nm–10 µm).

Aerosol number concentrations were compared via Wilcoxon rank-sum tests on paired data, with a Bonferroni adjusted p value for multiple comparisons. Speaking and breathing were assessed as the average number concentration of aerosol during the activity, whereas the peak number concentration was recorded for coughing. Given the non-parametric nature of the data, we report median and IQR for all results.

COVID-19 Patients were recruited on the infectious disease ward and had measurements of cough taken using the same methodology. Each measurement was taken at the bedside in single occupancy negative pressure rooms that draw clean air from the ventilation system above. However, background aerosol concentration was significantly higher than in the operating theatres, precluding reliable measurements of breathing and speaking.

RESULTS

Overall results and demographics

Thirty-three participants were recruited, of which 25 were healthy volunteers, and 8 were hospitalised patients with COVID-19. Thirteen (57%) of the volunteers were female, with a median age of 35 years (IQR 32–40 years), weight of 72 kg (IQR 64–79 kg), height of 1.74 m (IQR 1.64–1.79 m) and body mass index (BMI) of 23.6 kg/m^2 (IQR 22.0–25.5 kg/m^2).

Hospitalised patients with COVID-19 were older (mean age 55 years, IQR 49–59 years), with five men and three women. Height and weight were available for two patients: both were 170 cm tall; one weighed 85 kg (BMI: 29.4 kg), the other weighed 139 kg (BMI: 48.1 kg).

Volunteer aerosol emission

Table 1 describes the number of times each activity was performed, and on how many volunteers, alongside aerosol emission for each activity. The number of activities does not match the number of participants, as some volunteers (n=6) repeated the assessments on a different day to check repeatability, and some measurements were only performed on certain participants.

Correlation between the APS and OPS devices was high (r=0.98 unlogged, r=0.80 logged), despite the differing methodologies. Therefore, further analysis reports the APS figures in the text only, except where stated.

Figure 1A,B,C shows the aerosol number concentrations of each activity for volunteers, as reported by the APS (see online supplemental figure S4 for the OPS) and shows the clear variation in aerosol concentrations as well as the typical log-normal distribution. For baseline measurements, speaking produced more aerosol than breathing, and wearing an FRSM significantly reduced measured aerosol emission during both speaking (median 0.88 vs 0.03 particles/cm^3, p<0.0001) and coughing (median 1.52 vs 0.12 particles/cm^3, p<0.0001).

Continuous positive airway pressure

As shown in figure 1, aerosol emission sampled from participants receiving CPAP is greatly reduced compared with baseline measurements while breathing, speaking and coughing. Even with a large induced face mask air leak (>50 L/min), the aerosol emission measured over that leak during coughing was lower than in participants not receiving CPAP (0.12 vs 1.52 particles/cm^3; p<0.0001). At the filtered CPAP exit port, the aerosol emission was negligible and much reduced compared with those emitted during breathing, speaking or coughing in ambient room air (p<0.0001 for all comparisons).

Removal of the CPAP mask was associated with some aerosol emission, but this was significantly less than a cough in a healthy volunteer (peak of 0.36 particles/cm^3 vs 1.52 particles/cm^3, p<0.0001). In summary, CPAP was not associated with increased aerosolisation, but conversely was associated with much lower recorded aerosol number concentrations across all settings.

High-flow nasal oxygen

Assessment of aerosol emission from HFNO was complex. Our initial experiment used a single HFNO machine, with details described below.

HFNO was associated with increased aerosol number concentrations compared with breathing ambient room air (median aerosol in HFNO 30 L/min, 0.277 particles/cm^3; HFNO 60 L/min, 1.86 particles/cm^3; ambient air 0.03 particles/cm^3, p<0.0001 for all comparisons).

Higher flow rates (60 L/min) were associated with higher reported aerosol number concentrations than lower flow rates (30 L/min) for speaking (1.86 vs 0.246 particles/cm^3, p<0.001), breathing (1.86 vs 0.277 particles/cm^3, p<0.001), but not coughing (3.01 vs 2.96 particles/cm^3, p=0.002), nor coughing with a surgical face mask (0.63 vs 0.24 particles/cm^3, p=0.007),

Table 1 Aerosol emission produced across all activities in healthy volunteers

Oxygen delivery	Activity	Number of measurements	Aerosol emission (APS, particles/cm^3)*	Aerosol emission (OPS, particles/cm^3)*
Nil	Breathing	25	0.044 (0.022–0.08)	0.042 (0.023–0.125)
Nil	Speaking	25	0.088 (0.064–0.212)	0.121 (0.075–0.237)
Nil	Speaking with FRSM	23	0.03 (0.016–0.131)	0.038 (0.013–0.166)
Nil	Cough	25	1.52 (0.601–3.06)	2.14 (0.49–4.382)
Nil	Cough with FRSM	23	0.12 (0.06–0.555)	0.15 (0.06–0.57)
HFNO (60 L/min)	Breathing	20	1.861 (1.54–3.458)	2.921 (2.127–5.044)
HFNO (60 L/min)	Speaking	20	1.855 (1.201–2.359)	2.571 (1.65–3.255)
HFNO (60 L/min)	Cough	21	3.006 (2.597–5.525)	4.25 (3.011–6.41)
HFNO (60 L/min)	Cough with FRSM	10	0.63 (0.21–2.189)	0.75 (0.375–1.89)
CPAP at 15 mm Hg	Breathing sampling at area of greatest natural leak	20	0.013 (0.009–0.024)	0.012 (0.009–0.035)
CPAP at 15 mm Hg	Breathing sampling at exit port	20	0.002 (0–0.006)	0 (0–0.002)
CPAP at 15 mm Hg	Speaking sampling at exit port	8	0 (0–0.002)	0.001 (0–0.002)
CPAP at 15 mm Hg	Cough sampling at exit port	19	0.04 (0.01–0.06)	0.04 (0–0.105)
CPAP at 15 mm Hg	Cough sampling at leak	17	0.12 (0.06–0.72)	0.21 (0–0.99)
CPAP at 15 mm Hg	Removing CPAP mask	6	0.36 (0.195–0.57)	0.36 (0.27–0.6)

*This is the median IQR across individuals; average particles/cm^3/s for continuous activities, peak particles/cm^3 for sporadic activities.
APS, Aerodynamic Particle Sizer; FRSM, fluid-resistant surgical mask; HFNO, high-flow nasal oxygen; OPS, Optical Particle Sizer.

as both did not meet the Bonferroni corrected threshold (p=0.0004).

On review, the characteristics of the aerosol emissions during HFNO were not consistent with production of aerosol from the respiratory tract or mucosal surfaces, and aerosol was emitted even when the machine was unattached to the patient. We therefore performed a set of experiments to assess the source of this aerosol and their size distribution, with full experimental detail and results in the online supplemental appendix (see 'Aerosol Concentrations Generated by HFNO' and online supplemental figures S5–S8).

Importantly, we found that aerosol emission varied greatly among machines (two of four tested machines did not generate any aerosol), and that the size distribution of aerosol was not consistent with aerosol from the respiratory tract.

Patients with COVID-19 versus healthy volunteers

In total, eight patients were recruited with COVID-19. Demographics of these patients are recorded in online supplemental table S2. Measurement of aerosol concentrations generated by these patients was technically difficult due to the acute clinical environment, infection control requirements and the" room's higher background aerosol number concentration, necessitating high efficiency particulate air (HEPA) filtration to reduce this concentration and allow respiratory aerosol measurements. Four participants were on standard, low-flow nasal oxygen, while the others were breathing room air. For four participants, the background aerosol concentration in the room was higher than median concentrations generated by speaking and breathing measured in the ultra-clean theatres, so we only report the aerosol emission from coughing.

Figure 2 shows the aerosol emission recorded during coughing for both patients and volunteers. Compared with volunteers, patients with COVID-19 had higher aerosol emission when coughing (n=8; 10.5 vs 1.52 particles/cm^3, p=0.002) and when coughing wearing an FRSM, although due to low numbers neither met Bonferroni correction (n=3, 0.94 vs 0.12 particles/cm^3, p=0.03).

Importantly, the size distribution of aerosol particles in patients with COVID-19 was very similar to healthy volunteers (online supplemental figure S4). Breathing, speaking and coughing all generated aerosol particles in a log-normal size distribution with the peak in the 0.5–1 µm diameter range, consistent with previous reports of the size distribution of respiratory aerosol emissions.[11–14] This supports the use of healthy volunteers as proxies for patients infected with SARS-CoV-2.

Repeated measurements

For a subset of healthy volunteers (n=6), repeated measurements were made 1 month later. In total, there were 116 measurements repeated, (76 APS; 40 OPS). Correlation with the original measurement was moderate (r=0.71 on logged data), although this was driven by strong correlation in breathing (r=0.81), rather than speaking (r=0.17 on logged data) and coughing (r=0.38 on logged data) suggesting aerosol concentrations from breathing are relatively consistent for any individual recorded over a period of time, given the limitations inherent in the small numbers. In general, measurements on the second visit were marginally lower, which may represent slight differences in the experimental set-up.

Online supplemental figures S1 and S2 show these data, coloured by participant (S1) and activity (S2). Online supplemental figure S3 shows a Bland-Altman plot of this relationship.

DISCUSSION

Summary

This study comprehensively characterised the aerosol generation during standard CPAP and HFNO procedures, as compared with normal breathing, speaking and coughing. CPAP delivered

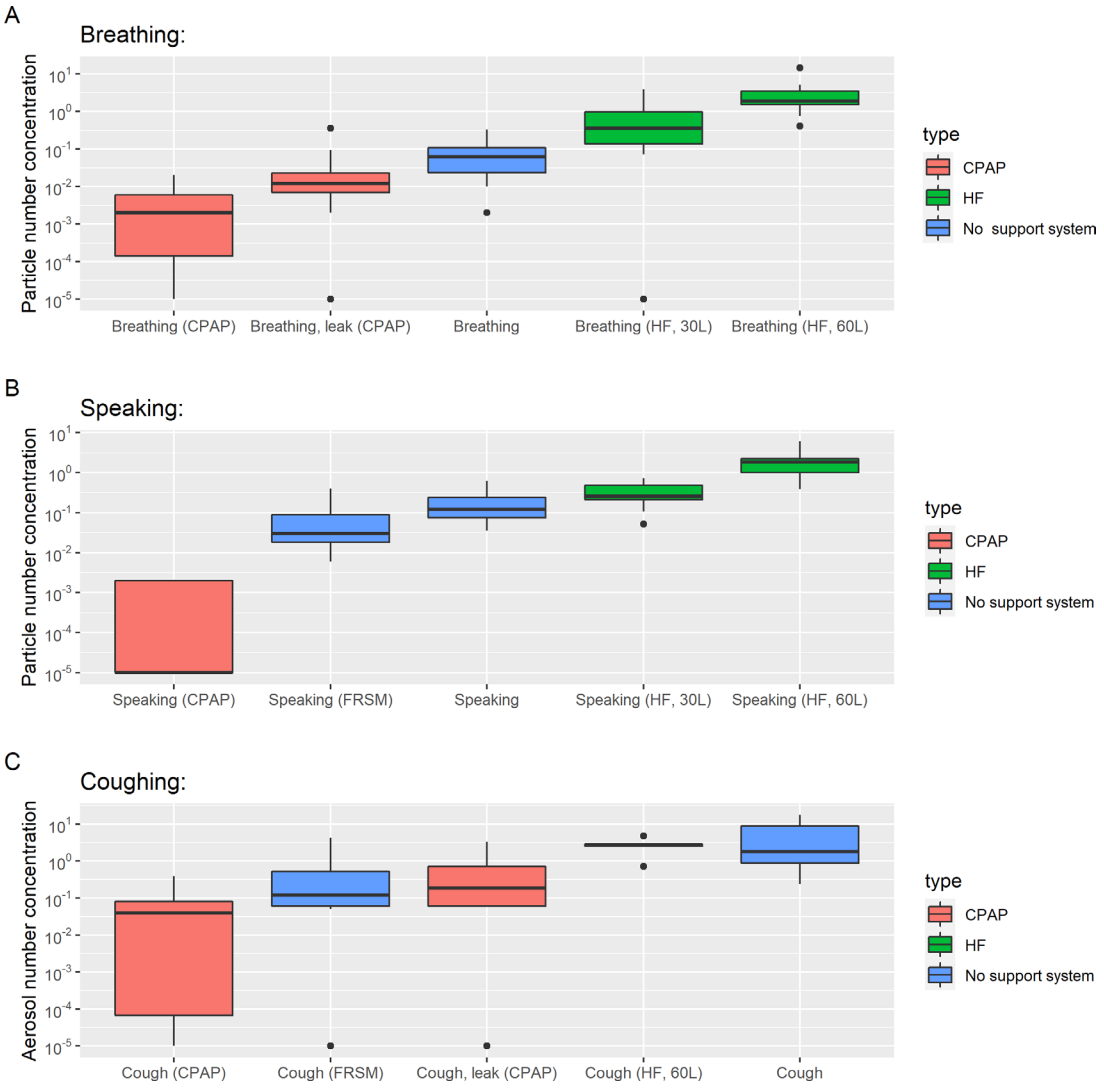


Figure 1 The aerosol number concentration sampled by an APS during baseline activities, CPAP or HFNO, reporting the mean concentration sampled during breathing (A) and speaking (B), and reporting the peak concentration sampled during coughs (C). Boxplots represent median and IQR. APS, Aerodynamic Particle Sizer; FRSM, fluid-resistant surgical mask; HFNO, high-flow nasal oxygen.

by face mask with exhalation filter was actually associated with lower aerosol number concentrations, even when large air leaks created around the CPAP face mask (>50 L/min) reflect the disruptions in CPAP of routine clinical care. For HFNO, aerosol concentrations were higher than baseline recordings. However, this additional aerosol was only present in some machines and largely disappeared with the use of a filter between the device and the patient. The size distribution of aerosol was unchanged when measured directly from the device or when attached to a patient, further supporting a non-biological origin.

Therefore, CPAP and HFNO should not be deemed AGPs, and provide no greater risk to healthcare staff relative to patients breathing, coughing and talking.

This is the first study to report on aerosol emission from patients with active COVID-19, with previous work on primates only.[15] While the data suggest that peak aerosol concentrations from coughs are higher than those from healthy volunteers without COVID-19, the background aerosol concentration on the ward was too high to report data on speaking and breathing.

Our analysis shows that a single cough generates at least 10-fold more aerosol particles at the peak concentration relative to the mean concentration for speaking or breathing (median concentrations of 1.52 particles/cm^3, 0.088 particles/cm^3 and 0.03 particles/cm^3 for cough, speaking and breathing, respectively, $p<0.0001$ for all comparisons).

In summary, our data (in concert with prior research on AGPs,[9 12] epidemiological studies showing lower risk to staff working in intensive care[16–20] and with viral loads higher earlier in infection, when patients are more often on the general wards[21]) suggest that risk of SARS-CoV-2 infection is not due to CPAP or HFNO generating infective aerosols. This has implications for infection and prevention control policy since aerosol generation appears greatest from patients with COVID-19 who are coughing.

Strengths and weaknesses

This study has multiple strengths. First, it was performed in ultra-clean laminar flow theatres, with very low aerosol background concentrations, allowing accurate quantification and attribution of aerosol emission. Second, the strong correlation between both aerosol measurement modalities provides confidence that the aerosol measurements are reliable. Third, repeated measurements and the recruitment of patients with active COVID-19

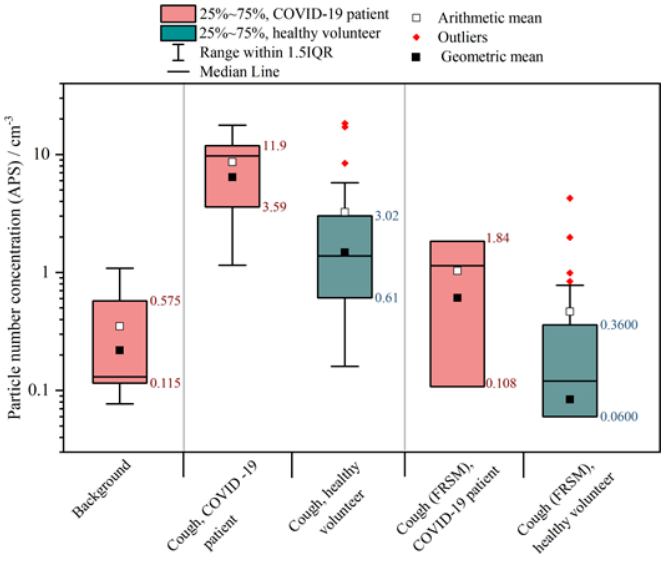


Figure 2 Box and whisker plot comparing the aerosol sampled by an APS when coughing in healthy subjects and by PCR-positive patients with COVID-19. APS, Aerodynamic Particle Sizer; FRSM, fluid-resistant surgical mask.

allow greater interpretation of clinical implications. Finally, the protocol reflects usual clinical care and is directly translatable to the health service delivery.

However, there are some important weaknesses. First, measurement methodology employed by the APS and OPS uses relatively low flow rates (1 L/min and 5 L/min, respectively). These mean that very short, high-impact aerosol emissions (eg, cough) may be hard to quantify. However, this applies to all APS and OPS technology, and does not limit relative comparison between oxygen delivery systems. Second, our assessment of patients with COVID-19 is limited, as we only recruited hospitalised patients and were only able to reduce background aerosol emission enough to reliably measure cough. Although these data are limited, they suggest that aerosol emission from patients with COVID-19 is likely to be higher than in volunteers, and underlines the difficulties in making these measurements in real patients.

It is important to note that the majority of our measurements (like in all other studies so far) come from healthy volunteers. It is likely that demographics (weight, height, age) have some impact on aerosol emission, and therefore some caution must be taken in extrapolating the raw data on aerosol emission, although we have no reason to suspect the changes in aerosol emission seen with delivery systems would dramatically change.

Finally, our non-humidified CPAP system uses full face masks with exhalation filters, which are standard care in our hospital and in many NHS hospitals outside critical care, following national policy at the start of the pandemic.[22] We cannot extrapolate to CPAP face masks without exhalation filters, although aerosol concentrations recorded from the face mask leak (unfiltered) were less than coughing without CPAP. As our system was unhumidified, we also cannot generalise to systems that use external humidification.

Similarly, the variability within the HFNO oxygen system used suggests that recorded aerosol emission may vary with device. As we only tested one manufacturer for each device, we cannot be sure that aerosol emission would differ with other manufacturers. However, as the mechanism of humidification and pressure generation is similar across different HFNO devices, it is likely that clinically relevant aerosol emission (eg, from the respiratory tract) is similar across devices. It is important to note that we cannot extrapolate to humidified CPAP devices, although we note the use of non-humidified CPAP is common in the management of patients with COVID-19 across many institutions.

We have chosen not to correct the reported particle concentrations sampled during each procedure to account for the effect of dilution by the airflow because the relative flow rate between each subject's different exhalation events compared with CPAP and HFNO is ill-defined. Thus, the uncorrected aerosol number concentrations as sampled by the APS and OPS do not represent the absolute quantity of particles generated by each activity, but can be used as a measure of the risk to a healthcare worker in the vicinity of the activity.

As activities such as coughing are forceful and short lived, these were analysed separately to the continuous activities (eg, breathing): short, transient activities are observed as a rapid rise in the reported number concentration followed by a decay over a few sample measurements (typically equivalent to 10–15 s for a cough) as the aerosol dissipates from the sampling funnel and is diluted by the clean room air. While reporting the intensive property of concentration allows us to compare relative yields from AGPs, it is important to note that estimating absolute yields or fluxes (extensive properties) requires knowledge of the volumetric flow rates for the gas in which the aerosol is dispersed. These present an additional challenge to measure. Although it is possible to report the absolute number of particles counted by the instruments (given we know their sampling volumetric flow rates), we cannot conclude that this is equivalent to the total aerosol yield without knowing the volumetric flow rate at aerosol source (ie, participant's mouth).

Comparisons with previous literature

There are few published studies of aerosol generation from oxygen delivery systems and respiratory support. The most similar study was performed by Gaeckle *et al*, which measured protocolised respiratory support systems in volunteers.[9] A similar protocol was used, although they also measured simple nasal cannulae and changes in respiration. Importantly, they reported a background aerosol concentration of ~0.060 particles/cm^3 (compared with zero under laminar flow), higher than we report for many activities (including breathing and speaking with an FRSM). As well as a high background, the aerosol number concentration was highly variable in their study (see figure 4 and E3 from reference 11, and figure 3 here for comparison). This variability makes reporting accurate aerosol concentrations for short events (eg, a cough) challenging, as we noted in recruiting our patients with COVID-19.

Consistent with our study, Gaeckle *et al* reported non-invasive ventilation to be non-aerosol generating. However, by contrast, they did not identify increased aerosol emission with HFNO. A very recent study, performed by Wilson *et al*,[23] measured aerosol counts in 10 healthy volunteers in a chamber, attempting to collect all exhaled aerosol. Similar to our study, they found coughing produced large amounts of aerosol compared with breathing and speaking. However, in contrast to our study, they identified small increases in aerosol emission with both CPAP (2.6-fold with single circuit) and HFNO (2.3-fold) with normal breathing. However, during exertion, they identified a reduction in aerosol emission with both of these therapies compared with breathing unaided. As Wilson *et al* comment (on our preprint), the differing results likely reflect different approaches to measuring and recording peak and average aerosol emission, but

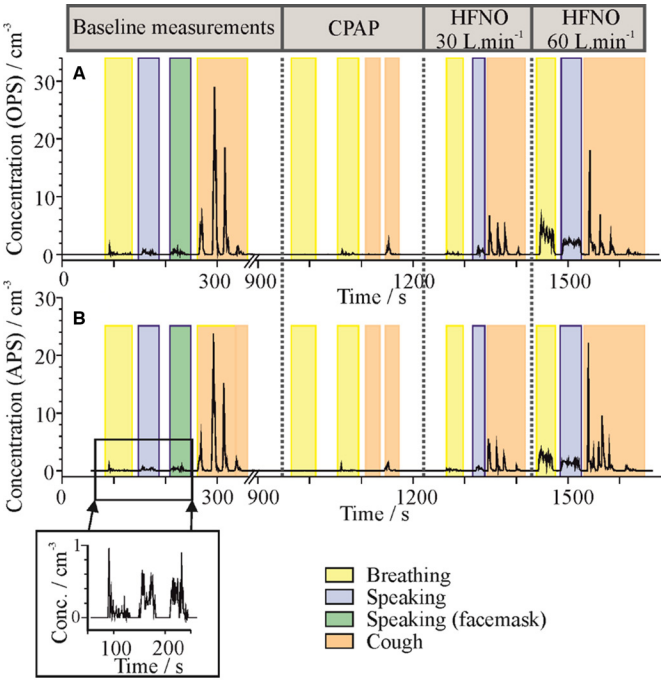


Figure 3 Example of the time series of OPS (A) and APS (B) number concentrations sampled during a measurement of one healthy subject performing baseline activities, followed by CPAP then HFNO. APS, Aerodynamic Particle Sizer; HFNO, high-flow nasal oxygen; OPS, Optical Particle Sizer.

the underlying results from both studies suggest that coughing represents a more significant source of aerosol than respiratory supports such as CPAP and HFNO.

The analysis presented in this paper, along with other work from our group identifying that intubation does not also generate significant aerosol,[12] suggest that the current infection and prevention control aerosol risk stratification strategies based on procedures rather than time spent in contact with patients coughing with COVID-19 may be misplaced.

Implications for clinical practice and policy

This study strongly supports re-evaluation of guidance removing CPAP as a high-risk AGP, with implications for more efficient delivery of NHS services. However, given that patients who receive acute respiratory support for COVID-19 are often acutely unwell and cough, the risk of aerosolisation of SARS-CoV-2 may be significant, complicating the policy changes. This work supports a re-evaluation of focusing solely on AGPs as potential risky events, and a shift towards focusing on the patient.

CONCLUSIONS

Non-humidified CPAP delivered via a filtered mask actually reduces aerosol emission compared with normal breathing. HFNO does generate additional aerosol, however this aerosol is generated from the machine and not the patient, and is unlikely to pose extra clinical risk given the size (<1 µm). Cough appears to generate significant aerosols in a size range compatible with airborne transmission of SARS-CoV-2. Policy around aerosol generation and infection control should be updated to reflect these adjusted risks.

Author affiliations
[1]Infection Science, North Bristol NHS Trust, Westbury on Trym, UK
[2]MRC Integrative Epidemiology Unit, Bristol, UK
[3]Bristol Aerosol Research Centre, School of Chemistry, University of Bristol, Bristol, UK
[4]Academic Respiratory Unit, North Bristol NHS Trust, Westbury on Trym, UK
[5]Physiotherapy Department, North Bristol NHS Trust, Westbury on Trym, UK
[6]Anaesthetics and Intensive Care Department, North Bristol NHS Trust, Westbury on Trym, UK
[7]Infectious Diseases, North Bristol NHS Trust, Bristol, UK
[8]Research and Development, North Bristol NHS Trust, Westbury on Trym, UK

Twitter Fergus W Hamilton @gushamilton and James William Dodd @theotherdodd

Collaborators The AERATOR group: Arnold D; Brown J; Bzdek BR; Davidson A; Dodd JW; Gormley M; Gregson F; Hamilton F; Maskell N; Murray J; Keller J; Pickering AE; Reid J; Sheikh S; Shrimpton A.

Contributors NAM, JWD, FWH, FKAG, DTA and JB designed the experiments. SS and FKAG analysed the data, with BRB and JPR providing supervisory support and analysis. EM coordinated inpatients with COVID-19, CW and AJM coordinated the study. KW provided expert opinion and analysis on respiratory support. JWD is the guarantor.

Funding NIHR-UKRI Rapid COVID Rolling Call (Ref: COV003). JWD's time was funded by MRC CARP Fellowship. (MR/T005114/1); FWH's time was funded by a GW4-CAT Wellcome Doctoral Fellowship; BRB's time was funded by an NERC grant (NE/P018459/1).

Competing interests None declared.

Patient consent for publication Not required.

Ethics approval This study was performed as part of the wider AERATOR Study to assess the risk of aerosolised transmission of SARS-CoV-2 in healthcare settings. Ethical approval was given by the North West Research Ethics Committee (Ref: 20/NW/0393, HRA approved 18/9/20).

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement All data relevant to the study are included in the article or uploaded as supplemental information. Anonymised aerosol data from the AERATOR Study will be submitted to the Bristol data repository (data.bris.ac.uk) on completion of the full study.

ORCID iDs
Fergus W Hamilton http://orcid.org/0000-0002-9760-4059
David T Arnold http://orcid.org/0000-0003-3158-7740
James William Dodd http://orcid.org/0000-0003-4805-5759

REFERENCES

1 Infection prevention and control during health care when novel coronavirus (nCoV) infection is suspected. Available: https://www.who.int/publications/i/item/10665-331495 [Accessed 1 Apr 2021].
2 Hamner L, Dubbel P, Capron I, *et al*. High SARS-CoV-2 Attack Rate Following Exposure at a Choir Practice - Skagit County, Washington, March 2020. *MMWR Morb Mortal Wkly Rep* 2020;69:606–10.
3 Liu Y, Ning Z, Chen Y, *et al*. Aerodynamic analysis of SARS-CoV-2 in two Wuhan hospitals. *Nature* 2020;582:557–60.
4 Klompas M, Baker MA, Rhee C. Airborne transmission of SARS-CoV-2: theoretical considerations and available evidence. *JAMA* 2020;324:441–2.
5 Public Health England. 6. COVID-19 infection prevention and control guidance: aerosol generating procedures, 2020. Available: https://www.gov.uk/government/publications/wuhan-novel-coronavirus-infection-prevention-and-control/covid-19-infection-prevention-and-control-guidance-aerosol-generating-procedures [Accessed 1 Apr 2021].
6 Public Health England. COVID-19: personal protective equipment use for aerosol generating procedures, 2020. Available: https://www.gov.uk/government/publications/covid-19-personal-protective-equipment-use-for-aerosol-generating-procedures [Accessed 24 Jun 2021].
7 World Health Organization. *Pandemic and epidemic diseases, world Health organization. infection prevention and control of Epidemic- and Pandemic-prone acute respiratory infections in health care: who guidelines*. Geneva: Pandemic and Epidemic Diseases, World Health Organization, 2014.
8 Klompas M, Baker M, Rhee C. What is an Aerosol-Generating procedure? *JAMA Surg* 2021;156:113–4.

9 Gaeckle NT, Lee J, Park Y, *et al*. Aerosol generation from the respiratory tract with various modes of oxygen delivery. *Am J Respir Crit Care Med* 2020;202:1115–24.
10 Hui DS, Chow BK, Lo T, *et al*. Exhaled air dispersion during high-flow nasal cannula therapy *versus* CPAP *via* different masks. *Eur Respir J* 2019;53. doi:10.1183/13993003.02339-2018. [Epub ahead of print: 11 04 2019].
11 Asadi S, Wexler AS, Cappa CD, *et al*. Aerosol emission and superemission during human speech increase with voice loudness. *Sci Rep* 2019;9:2348.
12 Brown J, Gregson FKA, Shrimpton A, *et al*. A quantitative evaluation of aerosol generation during tracheal intubation and extubation. *Anaesthesia* 2021;76:174–81.
13 Johnson GR, Morawska L, Ristovski ZD, *et al*. Modality of human expired aerosol size distributions. *J Aerosol Sci* 2011;42:839–51.
14 Alsved M, Matamis A, Bohlin R, *et al*. Exhaled respiratory particles during singing and talking. *Aerosol Sci Technol* 2020;54:1245–8.
15 Edwards DA, Ausiello D, Langer R. Exhaled aerosol increases with COVID-19 infection, and risk factors of disease symptom severity. *bioRxiv* 2020.
16 Jones CR, Hamilton FW, Thompson A, *et al*. SARS-CoV-2 IgG seroprevalence in healthcare workers and other staff at North Bristol NHS trust: a sociodemographic analysis. *J Infect* 2021;82:e24–7.
17 Eyre DW, Lumley SF, O'Donnell D, *et al*. Differential occupational risks to healthcare workers from SARS-CoV-2 observed during a prospective observational study. *Elife* 2020;9. doi:10.7554/eLife.60675. [Epub ahead of print: 21 08 2020].
18 Martin CA, Patel P, Goss C, *et al*. Demographic and occupational determinants of anti-SARS-CoV-2 IgG seropositivity in hospital staff. *J Public Health* 2020. doi:10.1093/pubmed/fdaa199. [Epub ahead of print: 16 Nov 2020].
19 Shah ASV, Wood R, Gribben C, *et al*. Risk of hospital admission with coronavirus disease 2019 in healthcare workers and their households: nationwide linkage cohort study. *BMJ* 2020;371:m3582.
20 Shields A, Faustini SE, Perez-Toledo M, *et al*. SARS-CoV-2 seroprevalence and asymptomatic viral carriage in healthcare workers: a cross-sectional study. *Thorax* 2020;75:1089–94.
21 Cevik M, Tate M, Lloyd O, *et al*. SARS-CoV-2, SARS-CoV, and MERS-CoV viral load dynamics, duration of viral shedding, and infectiousness: a systematic review and meta-analysis. *Lancet Microbe* 2021;2:e13–22.
22 England NHS. Guidance for the role and use of non-invasive respiratory support in adult patients with coronavirus (confirmed or suspected). Available: https://amhp.org.uk/app/uploads/2020/03/Guidance-Respiratory-Support.pdf
23 Wilson NM, Marks GB, Eckhardt A. The effect of respiratory activity, ventilatory therapy and facemasks on total aerosol emissions. *bioRxiv* 2021.

Aerosol emission from the respiratory tract: an analysis of aerosol generation from oxygen delivery systems.

SUPPLEMENTARY APPENDIX 1

Hamilton F*[1,2], Gregson F*[3], Arnold D[4], Sheikh S[3], Ward K[5], Brown J[6], Moran E[1],White C[7] AERATOR group, Bzdek BR[3], Reid JP[3], Maskell N[4#], Dodd JW[4#]

*These authors contributed equally

#These authors contributed equally

1. Infection Sciences, North Bristol NHS Trust, BS10 5NB
2. MRC Integrative Epidemiology Unit, University of Bristol, BS8 2HW
3. Bristol Aerosol Research Centre, School of Chemistry, University of Bristol, BS8 1TS
4. Academic Respiratory Unit, University of Bristol, BS10 5NB
5. Physiotherapy Department, North Bristol NHS Trust, BS10 5NB
6. Intensive Care Unit, North Bristol NHS Trust, BS10 5NB
7. Research and Innovation, North Bristol NHS Trust, BS10 5NB

AERATOR group (in alphabetical order): The AERATOR group consists of (in alphabetical order): Arnold, D; Brown, J; Bzdek, B; Davidson, A; Dodd, JW; Gormley M; Gregson, F; Hamilton, F; Maskell, N; Murray, J; Keller, J; Pickering, A.E; Reid, J; Sheikh, S; Shrimpton, A.

Corresponding author:

James Dodd (james.dodd@bristol.ac.uk)

Full Technical Methods

Study design

This was performed as part of the wider AERATOR study to assess the risk of aerosolised transmission of SARS-CoV-2 in healthcare settings. Ethical approval was given by the North West Research Ethics Committee (Ref: 20/NW/0393, HRA Approved 18/9/20).

Aerosol measurement

Aerosol measurements were recorded using two devices simultaneously: an Optical Particle Sizer (OPS) and Aerodynamic Particle Sizer (APS). Technical specifications were detailed in a previous publication from our group but are replicated here.[1] The key difference was the use of a shorter length of sampling tube (0.45m) from the sampling funnel through to the instrument inlet as patients were seated for this experiment, not supine.

The APS (TSI Incorporated, model 3321, Shoreview, NM, USA) measures aerosol at a sampling flow rate of 1 L min^{-1} with accompanying sheath flow of 4 L min^{-1}. The APS reports the aerodynamic size of particles in an aerosol plume, size-resolving aerosol number concentration into 52 size bins ranging from 0.5 µm to 20 µm in diameter with a time integration of 1 s. The size bins are equally spaced in log(diameter) space, apart from the smallest size bin (0.5 - 0.523 µm).

The OPS (TSI Incorporated, model 3330, Shoreview, NM, USA) samples air at 1 L min^{-1} and detects particles by laser optical scattering. The OPS reports the particle number concentration and optical size distribution within the diameter range 300 nm to 10 µm with a time resolution of 1 second. The OPS is widely used for aerosol studies from laboratories / clean rooms to more demanding outdoor environments. It is calibrated by the manufacturer using polystyrene latex spheres and its performance conforms to the ISO standard 21501-4:2018. The reported optical size of the particles is based on an assumed refractive index of pure water at 600 nm wavelength (1.333).

Both the APS and OPS were connected to the same sampling funnel, which was 3D printed (RAISE3D Pro2 Printer, 3DGBIRE, Chorley, UK) from PLA with a maximum diameter of 150 mm, cone height of 90 mm with a 10-mm exit port. Two conductive silicone sampling tubes of 0.3 m length and internal diameter 4.8 mm (3001788, TSI) were connected to the neck of the sampling funnel, with one connected to the APS and the other to the OPS.

For baseline measurements and HFNO, the funnel was placed such that the top of the funnel cone was 1 cm from the participant's forehead, and the sampling apex of the funnel was 10 cm from the

participant's mouth. For CPAP the sampling apex of the funnel was 10 cm from the exit port or area of greatest facemask leak. Supplement 1 includes some images from the set up to aid visualisation.

Environmental set up and patient recruitment

Observations were performed in two settings: healthy volunteers were recruited in an ultra-clean laminar flow operating theatre (EXFLOW 32, Howarth Air Technology, Farnworth, UK) with high efficiency particulate air (HEPA) filtration and an air supply rate of 1200 m^3/s. This ventilation system has a canopy 'clean zone' where surgical procedures are performed; the air circulation velocity is 0.2 $m.s^{-1}$ at 1 m above the floor below the canopy and produces 500–650 air changes per hour. All aerosol recordings were performed under the canopy, and the background aerosol concentration was sampled prior to each measurement for a mean sampling duration of 43 s, with mean background number concentrations of 0.00187 (SD 0.00271) cm^{-3} and 0.00330 (SD 0.00395) cm^{-3} reported by the APS and OPS, respectively. Air temperature in theatres was set to 20 °C and humidity between 40 and 60%.

The NIV machine used for delivering CPAP was the Phillips Trilogy 202 and the HFNO machine was the Fisher and Paykel Airvo 2. They were used according to manufacturer instructions. The CPAP circuits used were Armstrong Medical: AMVC1792/032 and Respironics: 1065830, CPAP full face masks were ResMed: AcuCare F1-0 and X, and the nasal cannula used were Fisher and Paykel Optiflow OPT944. The CPAP masks were unvented, as is standard UK practice, with a filtered exhalation valve. At the start of the pandemic, NHS England issued guidance recommending against external humidification, so our CPAP set up was unhumidified.[2]

Patients hospitalised with PCR-positive COVID-19 pneumonia were recruited and measurements taken in negative pressure ventilated side-rooms in the infectious disease ward. To reduce the background aerosol number concentration sufficient to allow measurements of baseline procedures in the ward setting, we used a portable HEPA filter (PUREAir R150, PUREAir Limited) with an airflow rate of 1500 m^3/hour for some patients. This reduced the background concentration for most measurements, although the average background number concentration was 0.351 (SD 0.382) cm^{-3} and 0.407 (SD 0.472) cm^{-3} for the APS and OPS, respectively, still significantly higher than in the laminar flow theatres. Where available, participant height and weight were recorded.

Healthy volunteers were recruited and invited to undergo a protocolised sequence of procedures in laminar flow environment of an operating theatre (see below for full procedural list). These included: tidal breathing, speaking (with/without mask), coughing (with/without mask), and

receiving CPAP (non humidified, full face mask, Continuous Positive Airway Pressure 15cmH_2O), and HFNO (High flow nasal oxygen). All procedures were performed in the seated position. For some participants, additional measurements were taken (e.g. speaking during HFNO, taking off the CPAP mask). Height and weight were recorded for all volunteers. A sample of the volunteers were invited to have a second measurement on a different date to ensure replicability. For CPAP, sampling was at the filtered exit port of the facemask, and at the point of maximum leak from the mask (measured by an experienced operator). If the maximum leak was less than 50 L/min, a leak was generated by the operator and measurement performed as close as possible to this leak. As with baseline measurements, speaking, breathing, and coughing were recorded during CPAP. Finally, measurements were made as the mask was removed for a small number of participants. CPAP settings were set to our hospital standard (15cm H_2O pressure), after initial scoping measurements and earlier research found no difference in aerosol emission with changing pressure, oxygen and humidity settings.

For HFNO, As with CPAP, measurements were made during tidal breathing, speaking, and coughing. For some participants, we tested the effect of wearing a surgical facemask over the HFNO nasal cannula on aerosol emission. As described in detail in supplement (Supplement S2), we also performed a set of measurements and experiments with four separate HFNO machines in order to identify the source of aerosol that we recorded during our study.

Hospitalised patients with COVID-19 were recruited by study members and had simple baseline measurements performed (e.g. speaking, breathing, and coughing, both with and without surgical facemasks). However, as background aerosol concentration was too high to reliably report aerosol emission from breathing and speaking, we only report aerosol emission from coughing.

Statistical analysis

Aerosol generation differs greatly among people, with an approximate log-normal distribution in number concentration.[3,4] As such, our analysis focussed on comparing the relative aerosol number concentrations from different procedures performed by each individual. We report the number concentration, an intensive property that does not depend on scale (i.e. is independent of the time or volume sampled) as reported by the instruments measured over a sample period, selected to be 1 s. We have reported one of two parameters for each activity: either the peak particle number concentration reported across the full number of samples of the measurement for single, forced exhalations such as coughing (cm^{-3}); or, the mean particle number concentration reported as the

average across all samples for continuous activities such as breathing or speaking (cm^{-3}). We then visualised size distributions of aerosol emission across the volunteers and compared aerosol emission across activities. As the data are non-parametric, we present median and interquartile ranges for results, and comparisons were made using the Wilcox-Sum rank test. Given the potential number of comparisons, we Bonferroni adjusted the p-value (0.05/120, p = 0.0004) to reduce false positive associations

Data analysis was performed by collating raw data of sampled aerosol concentration output by the APS and OPS instruments using Aerosol Instrument Manager 9.0 (TSI Incorporated, Shoreview, NM, USA) and Microsoft Excel. A custom-written software in LabVIEW (National Instruments, Texas, USA) was used to automate the analysis process for increased efficiency. For the PCR-positive hospitalised patients with COVID-19, the mean background aerosol number concentration was subtracted from the sampled aerosol number concentration for each activity to account for the non-zero background and to allow comparison with the data from healthy subjects collected under laminar flow. Formal statistical analysis was performed using R 4.0.3 (R foundation for Statistical Computing, Vienna). As a secondary objective, we wanted to test whether aerosol emission was different for patients with COVID-19 rather than healthy volunteers. For this, we compared activities in hospitalised patients with COVID-19 and in healthy controls. Finally, we tested a subset of volunteers twice, on different days and with different operators, to assess the intra-person variability in aerosol emission.

List of procedure, plan and set up:

Aerosol instruments on (baseline 30 s)	Measurement position
Breathe into funnel (30 s)	Funnel 10cm from mouth
STOP step back from funnel	
Speak into funnel (30 s) ~ 75 dB	Funnel 10cm from mouth
STOP step back from funnel	
Speak (30 s) **WITH FRSM** ~ 75 dB	Funnel 10cm from mouth

STOP step back from funnel	
Cough, then step back for 20 seconds	Funnel 10cm from mouth
Cough, then step back for 20 seconds	Funnel 10cm from mouth
Cough, then step back for 20 seconds	Funnel 10cm from mouth
Cough, **WITH FRSM**, then step back for 20 seconds	Funnel 10cm from mouth
STOP step back from funnel + place CPAP mask on	
Manouvere so CPAP exit port in funnel (30s) + breathing	CPAP exhalation port in funnel
STOP step back from funnel	
Manouvere so CPAP area of greatest leak in funnel (30s) + breathing	Funnel 10cm from area of greatest leak
STOP step back from funnel	
Manouvere so CPAP exit port in funnel (30s) + speaking	CPAP exhalation port in funnel
STOP step back from funnel	
Manouvere so CPAP area of greatest leak in funnel (30s) + speaking	Funnel 10cm from area of greatest leak
STOP step back from funnel	
Manouver so CPAP exit port in funnel, cough, then stand back 20 seconds	CPAP exhalation port in funnel
STOP step back from funnel	
Manouver so CPAP exit port in funnel, cough, then stand back 20 seconds	CPAP exhalation port in funnel
STOP step back from funnel	

Manouvere so CPAP area of greatest leak in funnel, cough, then stand back 20 seconds (30s)	Funnel 10cm from area of greatest leak
STOP step back from funnel	
Manouver so CPAP area of greatest leak in funnel, cough, then stand back 20 seconds	Funnel 10cm from area of greatest leak
STOP step back from funnel - remove CPAP mask	
Place NHF02 on at low flow with entrained air - measure tidal breathing (30s)	Funnel 10cm from mouth
STOP step back from funnel	
Place NHF02 on at low flow (20L) with entrained air - measure tidal breathing (30s)	Funnel 10cm from mouth
STOP step back from funnel	
Place NHF02 on at high flow (60L) with entrained air - measure tidal breathing (30s)	Funnel 10cm from mouth
STOP step back from funnel	
Place NHF02 on at high flow (60L) with entrained air - speaking (30s)	Funnel 10cm from mouth
STOP step back from funnel	
Place NHF02 on at high flow (60L) with entrained air - speaking (30s) + FRSM	Funnel 10cm from mouth
STOP step back from funnel	
Cough, then step back for 20 seconds (on 60L/min)	Funnel 10cm from mouth
Cough, then step back for 20 seconds (on 60L/min)	Funnel 10cm from mouth
Cough, then step back for 20 seconds (on 60L/min)	Funnel 10cm from mouth
STOP step back from funnel	

Note 1: Because coughs tend to decrease in strength, for some participants, we did a cough wearing a FRSM first in some participants.

Note 2: Speaking was asking patients to count from 1-100, at a set cadence and aiming for a similar volume between participants.

Images of the set up:

Figure S1: Line drawing of the set up in theatres from above. **A**: is the volunteer, **B** is the measuring equipment table, and **C** represents the limits of the laminar flow canopy.

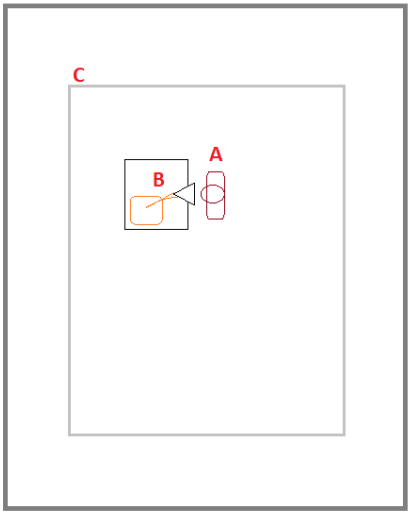


Figure S2: Image of the measuring equipment table, including **A: Funnel,** connected to **B: Silicon Tubing**, which connects to **C: Optical Particle Sizer** and **D: Aerodynamic Particle Sizer.**

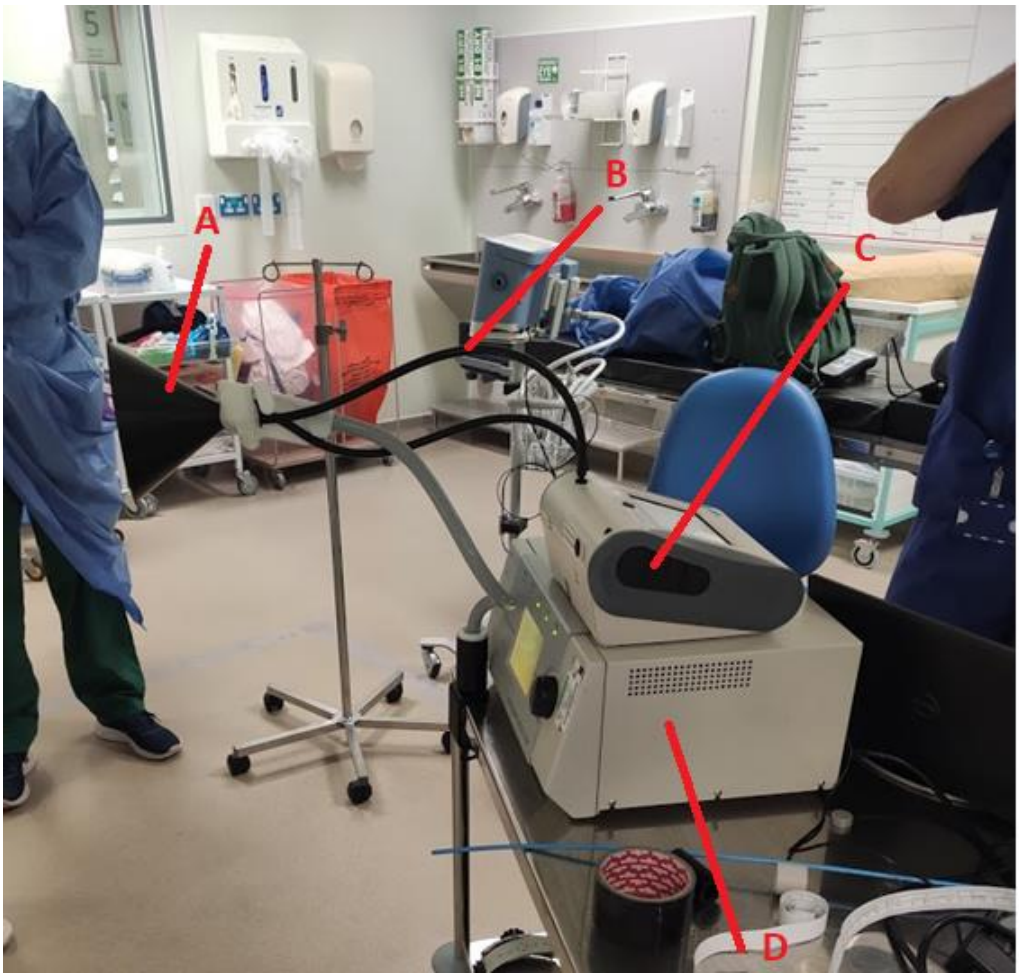


Figure S3: An example of someone receiving CPAP with the exit port facing the input funnel.

1. Brown J, Gregson FKA, Shrimpton A, et al. A quantitative evaluation of aerosol generation during tracheal intubation and extubation. Anaesthesia 2021;76(2):174–81.
2. England NHS. Guidance for the role and use of non-invasive respiratory support in adult patients with coronavirus (confirmed or suspected). Available from: https://amhp.org.uk/app/uploads/2020/03/Guidance-Respiratory-Support.pdf
3. Gregson, Watson, Orton, et al. Comparing the Respirable Aerosol Concentrations and Particle Size Distributions Generated by Singing, Speaking and Breathing. ChemRxiv [Internet] 2020 [cited 2021 Jun 24];Available from: https://chemrxiv.org/articles/preprint/Comparing_the_Respirable_Aerosol_Concentrations_and_Particle_Size_Distributions_Generated_by_Singing_Speaking_and_Breathing/12789221
4. Asadi S, Wexler AS, Cappa CD, Barreda S, Bouvier NM, Ristenpart WD. Aerosol emission and superemission during human speech increase with voice loudness. Sci Rep 2019;9(1):2348.

Aerosol emission from the respiratory tract: an analysis of aerosol generation from oxygen delivery systems.

SUPPLEMENTARY APPENDIX 2

Hamilton F*[1,2], Gregson F*[3], Arnold D[4], Sheikh S[3], Ward K[5], Brown J[6], Moran E[1],White C[7] AERATOR group, Bzdek BR[3], Reid JP[3], Maskell N[4#], Dodd JW[4#]

*These authors contributed equally

#These authors contributed equally

1. Infection Sciences, North Bristol NHS Trust, BS10 5NB
2. MRC Integrative Epidemiology Unit, University of Bristol, BS8 2HW
3. Bristol Aerosol Research Centre, School of Chemistry, University of Bristol, BS8 1TS
4. Academic Respiratory Unit, University of Bristol, BS10 5NB
5. Physiotherapy Department, North Bristol NHS Trust, BS10 5NB
6. Intensive Care Unit, North Bristol NHS Trust, BS10 5NB
7. Research and Innovation, North Bristol NHS Trust, BS10 5NB

AERATOR group (in alphabetical order): The AERATOR group consists of (in alphabetical order): Arnold, D; Brown, J; Bzdek, B; Davidson, A; Dodd, JW; Gormley M; Gregson, F; Hamilton, F; Maskell, N; Murray, J; Keller, J; Pickering, A.E; Reid, J; Sheikh, S; Shrimpton, A.

Corresponding author:

James Dodd (james.dodd@bristol.ac.uk)

Repeat Measurement Data

A subgroup of the healthy subjects (*n*=6) were invited to repeat the baseline measurements one month later. The comparison between aerosol concentrations sampled during the original measurement and later measurement are shown in Figure S4 and S5, showing the correlation between the two measurements (r = 0.71 on logged data).

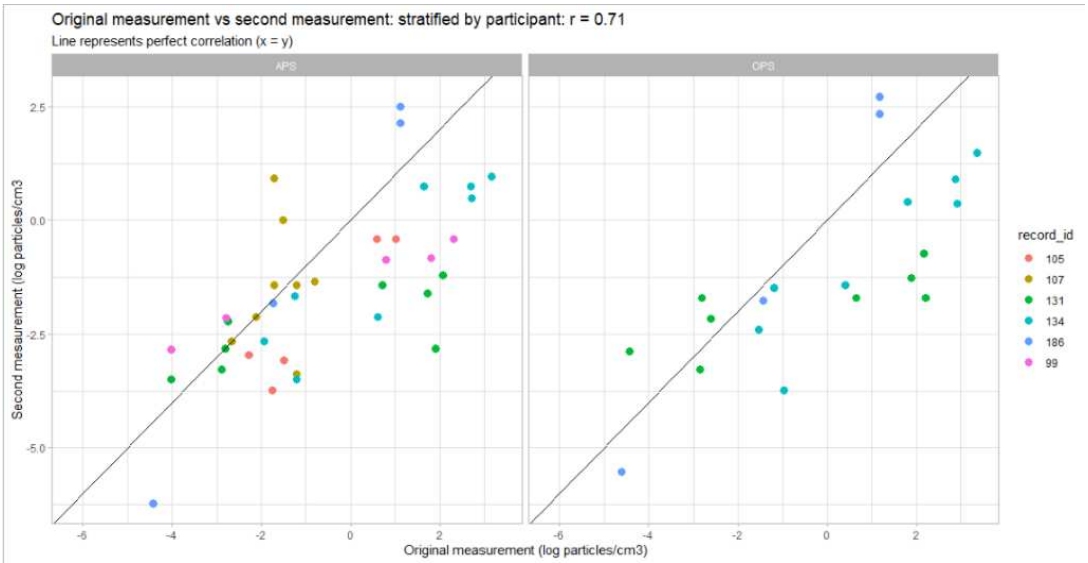


Figure S4: The aerosol number concentration sampled by an APS during baseline measurements for six subjects compared to that for a repeat measurement at a later date. For breathing and speaking activities, the mean concentration per sample (equivalent to 1 s acquisition) is reported; for coughing the peak concentration sampled during any sample (1 s acquisition) is reported. Data are classified by colour according to the subject (displayed in legend).

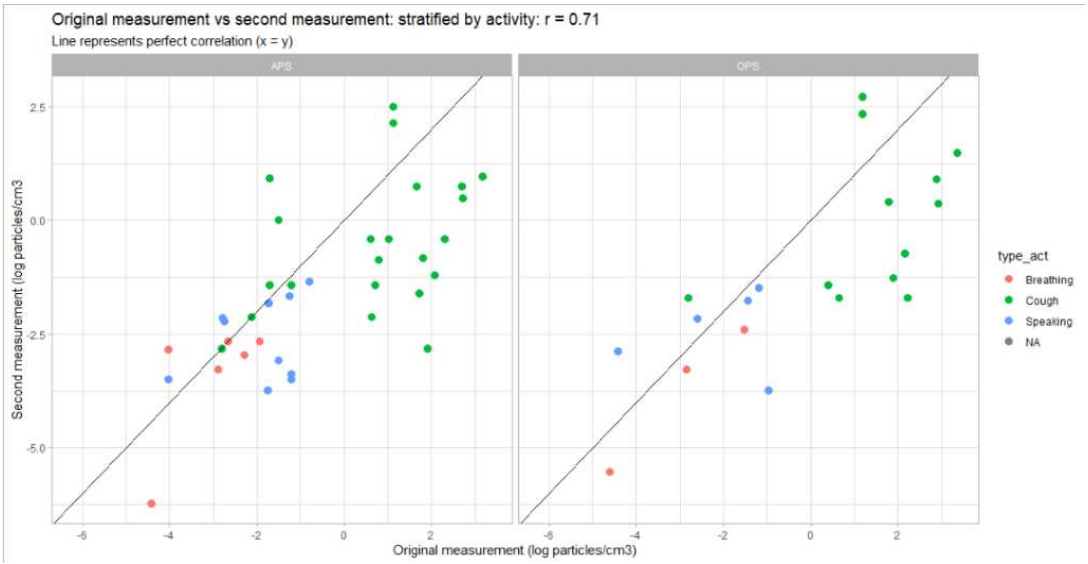
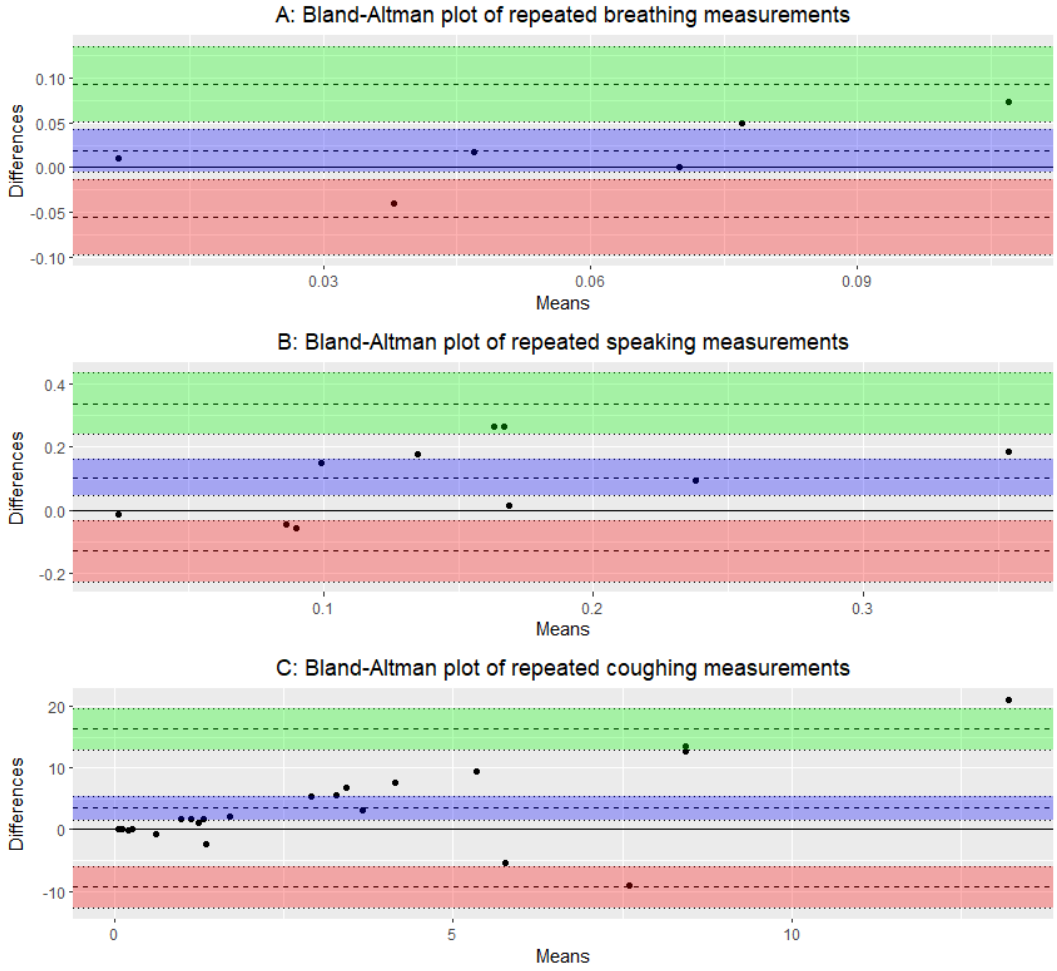


Figure S5: The aerosol number concentration sampled by an APS during baseline measurements for six subjects compared to that for a repeat measurement at a later date. For breathing and speaking activities, the mean concentration per sample (equivalent to 1 s acquisition) is reported; for coughing the peak concentration observed during any sample (1 s acquisition) is reported. Data is classified by colour according to the type of respiratory emission (displayed in legend).

Figure S6: A Bland-Altman plot of differences for Breathing (A), Speaking (B), and Coughing (C), from the APS. The dashed lines represent the difference and the 95% confidence interval point estimates, with the shading representing the confidence interval for each point estimate.



OPS Measurement Data

Aerosol concentrations were recorded simultaneously with an OPS and APS. The box and whisker plots for the OPS measurements (equivalent to Figure 1) are shown in Figure S7.

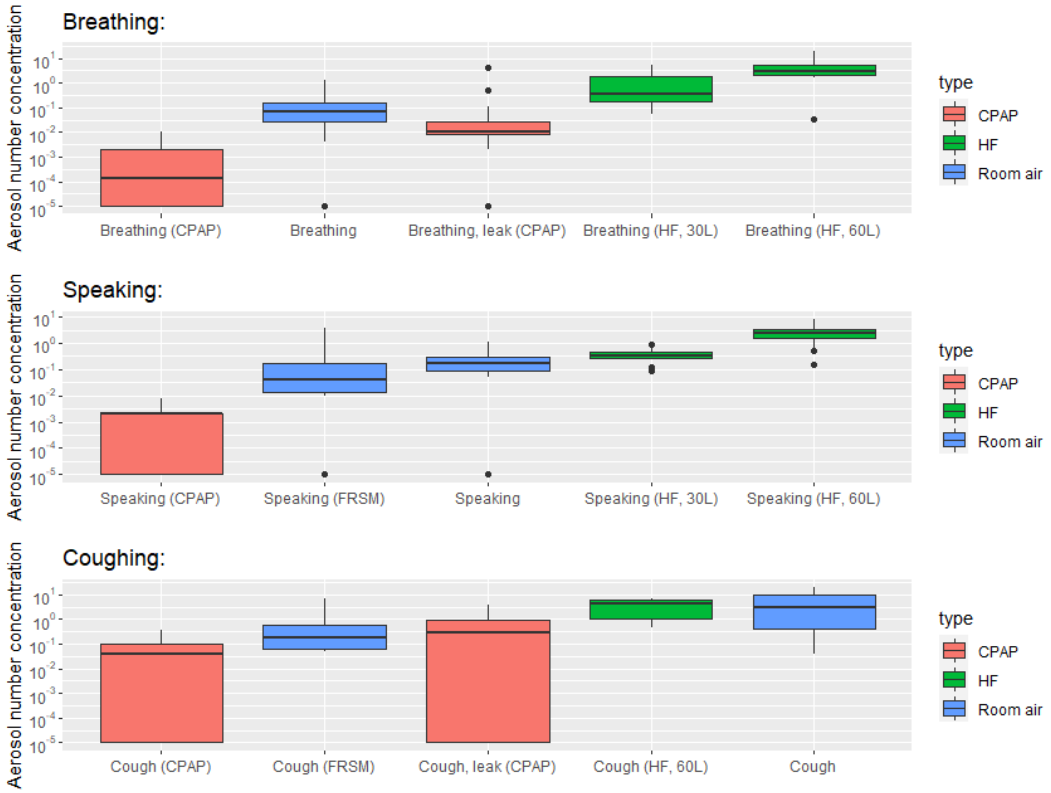


Figure S7: The aerosol number concentration sampled by an OPS during breathing, speaking, and coughing while under each activity recorded from the OPS, including a) the mean concentration sampled during breathing and speaking and b) the peak concentration sampled during coughs.

Size Distributions of Sampled Aerosol

The mean size distributions sampled by the APS from all subjects during different measurements are shown in Figure S8. The respiratory aerosol particles form a lognormal size distribution, with the majority of particles generated in the sub-micrometre diameter range for coughing, breathing and speaking. The aerosol particles sampled when a subject spoke or coughed while wearing a fluid-resistant surgical facemask had a reduced number concentration compared to when wearing no mask (0.113 vs 0.038, $p = 0.002$ and 1.40 vs 0.075, $p < 0.001$). This is particularly evident when looking at the size distributions in the diameter range 0.5 - 1.5 µm (Fig. S8). The particles generated by a cough during CPAP, sampled at the area of greatest leak (orange squares) appear to show a similar size distribution to a regular baseline cough (red squares), but are reduced in number concentration.

The mean size distribution of particles generated by coughs from patients infected with COVID-19 is also compared with that from healthy volunteers in Fig. S8. The sample size of the COVID-19 patients is reduced (n =7) and only shows data for which the aerosol peak could

be observed above the high background aerosol concentration, so the number concentration appears greater than that for healthy subjects. However, the size distribution of the sampled particles is similar, with the peak of the lognormal distributions for each activity aligning at a comparable aerodynamic diameter for healthy subjects vs. COVID-19 patients.

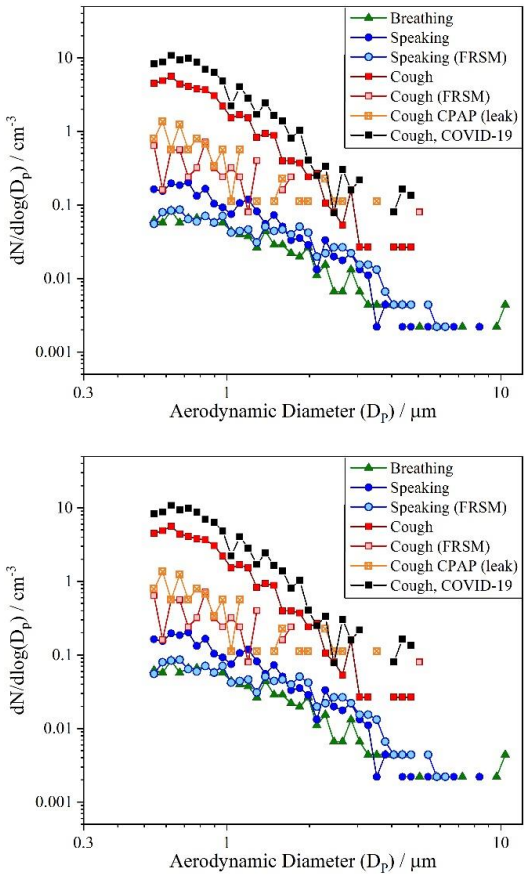


Figure S8: The mean size distribution of baseline measurements of breathing, speaking, speaking with a face mask, coughing, coughing with a facemask (all n=25), coughing during CPAP, sampling at the area of greatest leak (n = 17), and coughing by PCR-positive patients with COVID-19 (n=7).

Aerosol Concentrations Generated by HFNO

Considering the high aerosol level for all baseline activities conducted with 60 L min^{-1} HFNO compared to CPAP or no oxygen delivery system (Figure 1), the aerosol source from HFNO, both with and without a subject present, was investigated. One subject generated a mean aerosol concentration (sampled by an APS) during baseline speaking of 0.162 cm^{-3}, which increased to 1.7 cm^{-3} when speaking during HFNO (Figure S9). Given that the mean concentration sampled when the HFNO cannula with 60 L min^{-1} humidified air (no subject present) was held 10 cm from the sampling funnel apex was 1.312 cm^{-3} (Fig. S9), it appears that the additional particles sampled during the subject's measurement of speaking during

HFNO have originated from the HFNO device and were sampled in addition to the respiratory aerosol from speaking. Likewise, the mean baseline breathing aerosol concentration for this subject was 0.002 cm^{-3}, which rose to 2.4 cm^{-3} when the patient breathed during HFNO.

Introducing a 0-particle HEPA capsule filter (removal rating: 1.2 mm Versapor® membrane filteration area: 860 cm^2 , set-up shown in Figure S10 in series with HFNO tubing between the humidifier and the nasal cannula reduced the concentration of sampled aerosol from the HFNO device alone (no subject present) by two orders of magnitude to 0.005 cm^{-3}. When the subject was re-introduced to undergo HFNO, with the HEPA filter still present in series, the mean particle number concentration sampled was 0.006 cm^{-3} during the subject's breathing, three orders of magnitude lower than that with no HEPA filter inserted in the HFNO tubing. Therefore, it can be concluded that a large proportion of particles detected from HFNO activities are generated by the HFNO air flow rather than the subject. These trends were also observed in the sampled OPS concentration data (not shown).

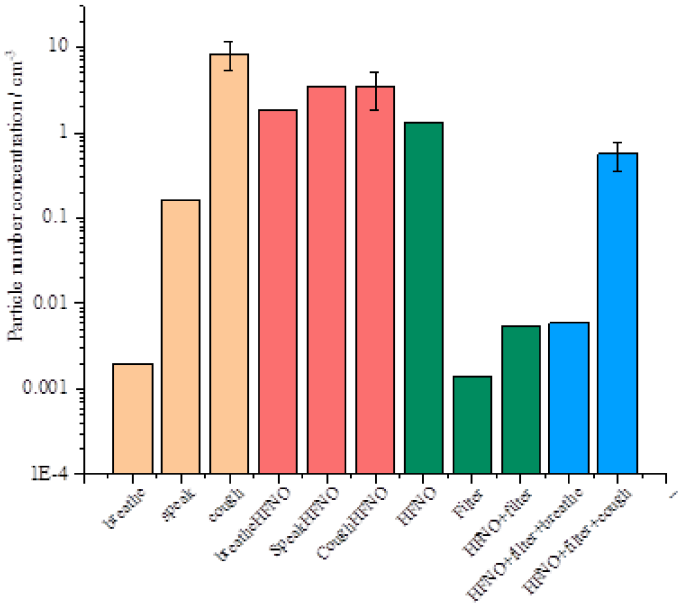


Figure S9: APS total particle count detected from 1 second of activity / cm-3 for breathing and speaking and peak height cough data / cm-3 from one subject assessing particle source during HFNO delivery. Baseline measurements (cream) show breathing and speaking and coughing data without HFNO. Baseline measurements whilst receiving HFNO delivery (coral) for subject are greater. Addition of the 0-particle HEPA filter without the subject present measurements are shown in green. The subject was then introduced with the filter in series (blue). The sustained, timed activities (breathing, speaking, HFNO device alone) have the mean concentration per sample reported (equivalent to 1 s acquisition) and concentrations produced during a cough are reported as the peak concentration observed during any one sample (1 s acquisition). Error shows SD.

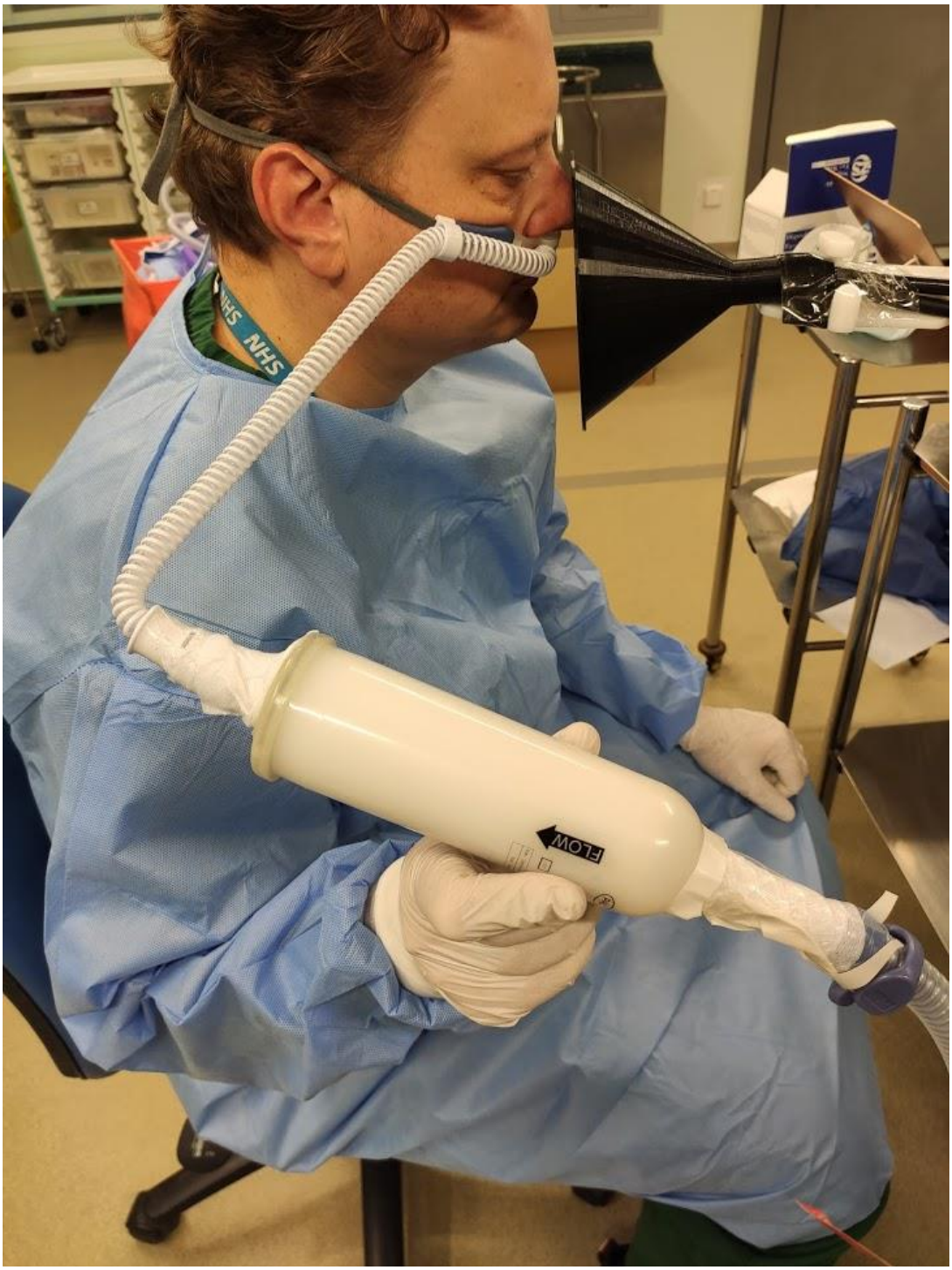


Figure S10: Experimental set up showing the insertion of the HEPA capsule filter into the tubing between the HFNO cannula and the HFNO device, sampled by the APS and OPS via the funnel. Please note, this does not describe the experimental set up used for recruitment of the healthy volunteers, where the volunteer would be further from the cone, but shows the zero particle filter between the HFNO device and the volunteer.

In order to further investigate the particle source from HFNO, a subject was invited to repeat the baseline measurements during HFNO but both with and without the HFNO humidifier in operation (Figure S11). There was no appreciable difference between the total particle concentration generated during this subject's baseline measurements during HFNO with the humidifier in operation or during dry HFNO. Speaking whilst receiving humidified HFNO generated a mean concentration of 0.844 cm^{-3}, compared to 0.872 cm^{-3} during dry HFNO. Whilst this additional experiment was performed for only one subject, the results indicate that the HFNO humidifier was not the major source of the particles generated from the HFNO delivery system.

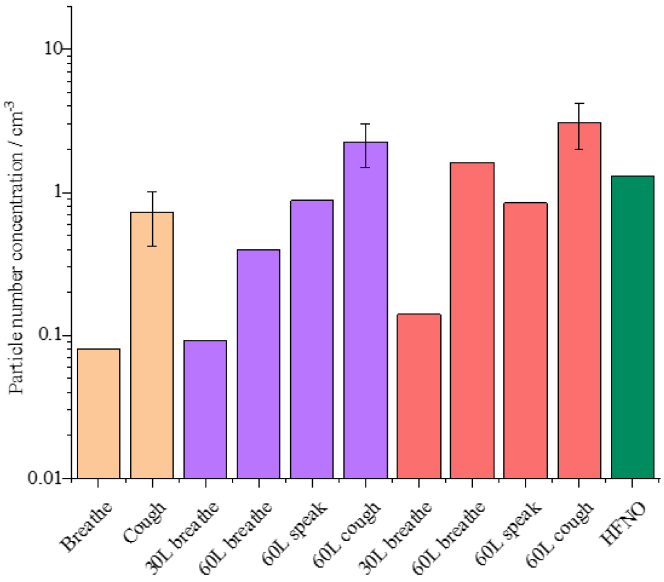


Figure S11: The mean concentration sampled by an APS during a subject's baseline measurements (cream), compared to that during dried HFNO (purple) and humidified HFNO (coral). The concentration of particles generated by the HFNO device alone (no subject present) is shown in green. The sustained, timed activities (breathing and speaking) have the mean concentration per sample reported (equivalent to 1 s acquisition) and concentrations produced during a cough are reported as the peak concentration observed during any one sample (1 s acquisition). Error shows SD.

Size Distributions of Aerosol Concentrations Generated by HFNO

Investigating the size distribution of particles sampled during subjects performing HFNO can also allude to their origin. Particles generated by the HFNO device alone and sampled by the OPS and APS are shown in Figure S12 (black squares and grey diamonds, respectively). When the subjects (*n*=25) undergoing HFNO breathed into the sampling funnel, the size distribution of particles sampled by the APS (green squares) comprises both those particles sampled directly from the HFNO device (HFNO device #1) as well as those sampled during baseline

breathing with no oxygen delivery (dark green triangles). Essentially, during HFNO breathing there appears to be an additive effect of the two populations of particles, both respiratory aerosol and aerosol generated by the HFNO device. The latter, whilst contributing to the nominal increase in particles sampled during HFNO breathing compared to baseline breathing, are not expected to carry COVID-19 transmission risk. As the HFNO device particles are very small in diameter (the peak of the size distribution is well below 0.5 μm and is near zero around 1 μm) these particles are expected to only follow the airflow streamlines through the nasal cavity and be immediately exhaled into the sampled funnel without impaction in the nasal canal or respiratory tract and thus cannot pick up any viral load during transport.

For a subset of the cohort (*n*=6), an additional measurement of breathing during HFNO was performed but with a different HFNO machine (HFNO device #4). The concentration and size distribution of particles generated by this HFNO device alone (no subject present) is different to the first HFNO device, and is shown in the inset to Fig. S12a. When the six subjects undergoing HFNO with the second device breathed into the funnel, there is clearly an additive effect of the two size distributions arising from the HFNO and the breathing.
This additive effect of sampling both the HFNO-generated particles and respiratory particles can be observed for speaking, with both the original HFNO device (*n*=25, Figure S12a) and with the second HFNO device (*n*=6, Figure S12a inset). This serves as evidence that whilst a greater particle concentration is sampled during HFNO compared to baseline measurement with no oxygen support, these additional particles arise from the HFNO device itself and do not carry an associated COVID-19 transmission risk.

The quantity and size distribution of particles generated by the HFNO device is variable across different machines. Figure S12d shows the OPS size distributions of particles originating from four HFNO devices. OPS size distributions are preferentially shown here because the particles are of small diameter that would not be detected by the APS. “HFNO 1”, used in the majority of the HFNO subject data (*n*=23, principal figures in Fig.S8), generated a mean OPS particle concentration of 1.622 cm^{-3}, “HFNO 4”, used in the additional HFNO cohort (*n*=6, in the insets of Fig. S12) generated a mean concentration of 3.45 cm^{-3}. The cleaner machines “HFNO 2” and “HFNO 3” generated fewer particles, 0.0070 cm^{-3} and 0.020 cm^{-3}, respectively, however these devices were not used for a cohort of subjects undergoing HFNO.

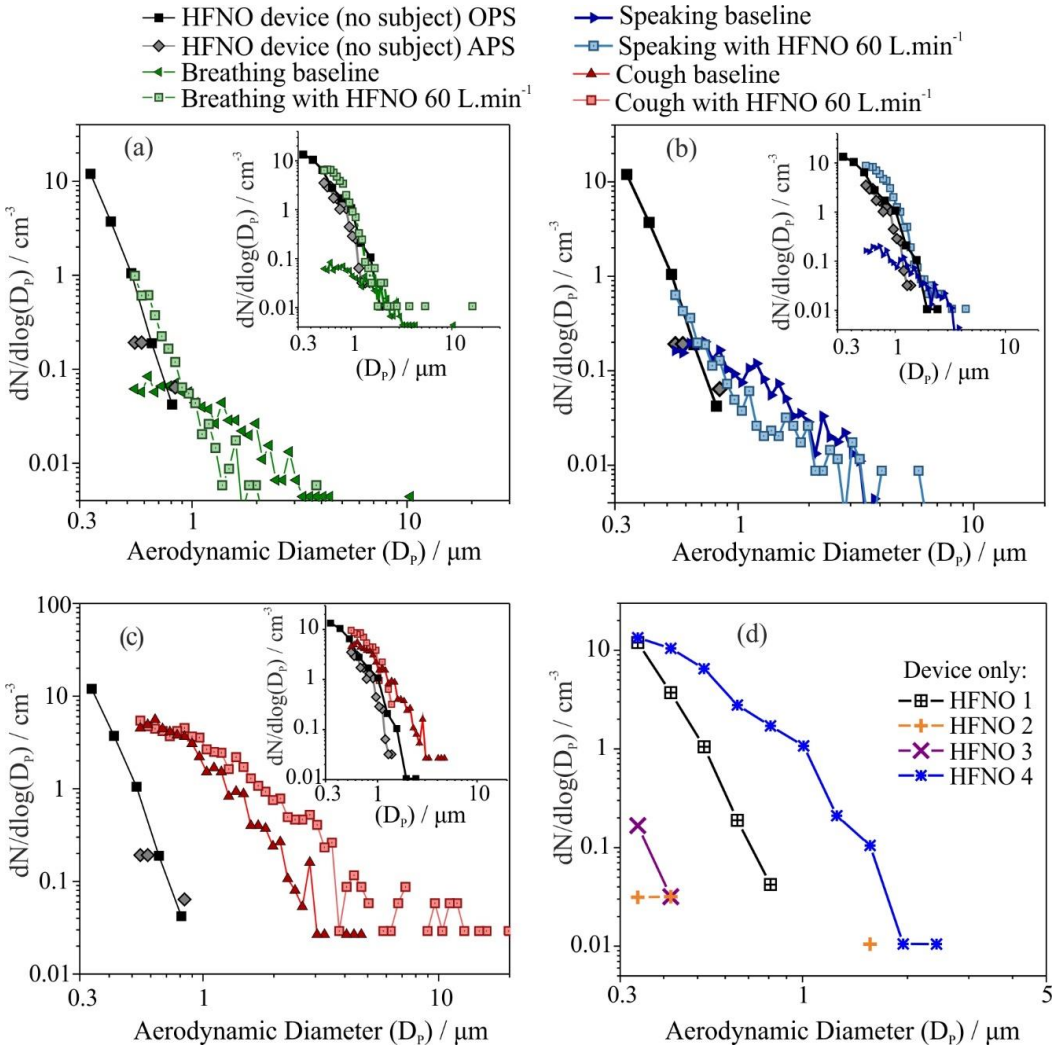


Figure S12: The mean size distribution of healthy subjects performing a baseline respiratory activity (*n*=25) and also during 60 L.min^{-1} HFNO (*n*=23), compared to the size distribution of particles generated by the HFNO device and cannula. Inset shows the mean distribution of healthy subjects performing the same baseline activity (*n*=6), but with an alternative HFNO device that generates more particles in a different size distribution. Respiratory activities are breathing, speaking and coughing for a), b) and c), respectively. d) OPS size distributions of particles originating just from the HFNO device for four different devices, where "HFNO 1" is that used for the majority of HFNO datasets (*n*=23) and "HFNO 4" is that used for the additional six subjects in the figure insets. HFNO devices 2 and 3 generate fewer particles but were not used for subject datasets.

The size distribution of particles sampled from a cough during HFNO do not show quite the same additive effect as those for breathing and speaking (Figure S12c and inset). The reasons behind this are unclear. Given that coughs are transient and short lived, different to the sustained respiratory activities of breathing and speaking, there are measurement challenges associated with quantifying the generated concentrations and so the trends of how the size distribution changes for a cough during HFNO may be more complex.

Demographics of COVID-19 positive patients:

Table S1: Demographics of COVID-19 patients.

Age (yrs)	Sex	Weight	Height	BMI	Ethnicity
46	Male			Not stated	Black or Black British - African
60	Male			Not stated	Not stated
50	Male			Not stated	Not stated
57	Female			Not stated	White- British
55	Male			Not stated	Not stated
48	Male	85kg	170cm	29.411	White- British
53	Female			Not stated	Not stated
60	Male	139kg	170cm	48.096	Not stated