



Original Article

Comparison of virus aerosol concentrations across a face shield worn on a healthcare personnel during a simulated patient cough

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Abstract

Background: Patients diagnosed with coronavirus disease 2019 (COVID-19) aerosolize severe acute respiratory coronavirus virus 2 (SARS-CoV-2) via respiratory efforts, expose, and possibly infect healthcare personnel (HCP). To prevent transmission of SARS-CoV-2 HCP have been required to wear personal protective equipment (PPE) during patient care. Early in the COVID-19 pandemic, face shields were used as an approach to control HCP exposure to SARS-CoV-2, including eye protection.

Methods: An MS2 bacteriophage was used as a surrogate for SARS-CoV-2 and was aerosolized using a coughing machine. A simulated HCP wearing a disposable plastic face shield was placed 0.41 m (16 inches) away from the coughing machine. The aerosolized virus was sampled using SKC biosamplers on the inside (near the mouth of the simulated HCP) and the outside of the face shield. The aerosolized virus collected by the SKC Biosampler was analyzed using a viability assay. Optical particle counters (OPCs) were placed next to the biosamplers to measure the particle concentration.

Results: There was a statistically significant reduction ($P < .0006$) in viable virus concentration on the inside of the face shield compared to the outside of the face shield. The particle concentration was significantly lower on the inside of the face shield compared to the outside of the face shield for 12 of the 16 particle sizes measured ($P < .05$).

Conclusions: Reductions in virus and particle concentrations were observed on the inside of the face shield; however, viable virus was measured on the inside of the face shield, in the breathing zone of the HCP. Therefore, other exposure control methods need to be used to prevent transmission from virus aerosol.

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Severe acute respiratory coronavirus virus 2 (SARS-CoV-2) is a positive sense, single-stranded, RNA virus in the coronavirus family. The virus was originally detected in humans in 2019 and has since caused a global pandemic with >533 million cases and 6.3 million deaths as of June 2022. SARS-CoV-2 causes coronavirus disease 2019 (COVID-19). The disease has symptoms including fever, cough, shortness of breath, fatigue, sore throat, nausea, and diarrhea. In the United States, ~80 million patients have been hospitalized with COVID-19.

There may be a dose response associated with COVID-19 severity,¹ meaning that the dose of virus in the initial inoculum could affect the severity of disease.¹ Patients can aerosolize virus, which exposes the HCP. Therefore, reducing SARS-CoV-2 aerosol concentration and worker exposure could reduce the disease burden among HCPs. Virus containing aerosols can be reduced by

following the industrial hygiene hierarchy of controls, which provides guidance on reducing exposure to hazards through elimination, substitution, engineering controls, administrative controls, or personal protective equipment (PPE). PPE is often used in healthcare to reduce exposure to viruses and can be worn by the HCP.² PPE can include gowns, gloves, face shields, goggles, masks, and respirators. Face shields have been used in the past and are currently being used as PPE to reduce exposure to bioaerosols and as a barrier device for eye protection.² During the COVID-19 pandemic, 70% of US hospitals reported requiring eye protection.³ Thus, it is crucial that we understand the effectiveness of a disposable face shield at reducing virus containing aerosol concentrations in the breathing zone of an HCP when they are exposed to coughing patients as part of effectively protecting workers in a high-risk environment.

Coughing machines have been previously developed to study bioaerosols and particles produced when a human coughs.^{4,5} We developed a coughing machine to study the effectiveness of a disposable plastic shield at reducing HCP exposure to SARS-CoV-2 via aerosol. MS2 bacteriophage has been used as a safe

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surrogate for the aerosol transmission of virus.^{6–9} MS2 is a single-stranded RNA bacteriophage that is nonenveloped.²

Prior studies with coughing machines have not analyzed the effectiveness of PPE with viable virus. Therefore, in the experiment performed in our study, we compared SARS-CoV-2 aerosol particle concentrations, through the surrogate MS2, across a face shield during a simulated cough. We analyzed virus concentrations and viability across a disposable plastic face shield placed on a simulated HCP during a simulated cough from a simulated patient and performed statistical analyses to compare the virus concentrations found across the face shield. We used these data to analyze the effectiveness of the face shield at reducing virus exposure.

Materials and methods

Power analysis

A power analysis was conducted a priori, with an effect size of 1, to determine the sample size needed to result in 80% power when using a paired *t* test with an α set at the 0.05 using a normal distribution. We performed the calculations using R Core Team 2020 (R: A language and environment for statistical computing, R Foundation for Statistical Computing, Vienna, Austria). The determined sample size was 16.7 for each group; therefore, at minimum, 17 trials were conducted.

Coughing machine virus aerosolization

A coughing machine was designed to simulate a patient coughing viral aerosolized particles and exposing HCPs (article currently under review). The coughing machine operates by pushing 4.1 L of air, which is mixed with aerosolized virus through a mannequin's mouth, producing a coughing plume. The resulting plume is like a human cough. A simulated HCP was placed 0.41 m in front of the cough to simulate exposure in a hospital or other care facility. A face shield was placed on the simulated HCP to mimic part of the PPE worn. MS2, the bacteriophage used as a surrogate for SARS-CoV-2 was placed in the liquid reservoir of the coughing machine and was aerosolized during the cough. Three consecutive simulated coughs were performed in each trial to mimic a "coughing fit."

Virus and particle sampling

Virus and particle concentrations were sampled across the disposable plastic face shield (placed on a simulated HCP) while the coughing machine was operated. Two SKC biosamplers (no. 225-9595, SKC Inc, Eighty-Four, PA) were used to sample the MS2 (virus concentration) and 2 optical particle counters (OPCs) (GRIMM 11-D Aerosol Counters, nos. 619012 and 619025, Durag Group, Germany) were used to sample the particle concentration. The Biosamplers and OPCs were placed on the inside and the outside of the simulated HCP face shield (Fig. 1). The liquid media used in the biosampler was 20 mL phosphate-buffered saline (PBS, Gibco, 1x, pH 7.4 +/-, TX). The biosamplers were each connected to a pump that sampled at 12.5 liters per minute (LPM). The OPCs used a 1-second interval and sampled number concentration information across 16 channel (bin) sizes (range, 0.253–3.515 μm). The biosamplers and OPCs were started 1 minute prior to the first cough and operated nonstop throughout the 3 coughs (~28 seconds).

The sampling was conducted in an undisturbed chamber (8.5 m \times 3.7 m \times 2.4 m). Relative humidity and temperature were

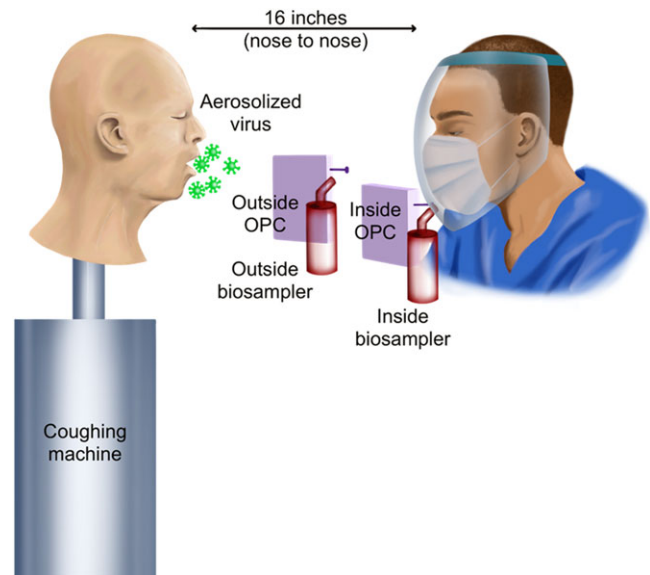


Figure 1. Sampling setup. The disposable plastic face shield is placed on the simulated healthcare personnel with the coughing mannequin representing a patient with COVID-19. Biosamplers and optical particle counters (OPCs) are placed on the inside and the outside of the face shield to evaluate the healthcare personnel's exposure to bioaerosols produced by coughing.

also monitored to ensure consistency across trials using a direct reading instrument (Extech Instruments Hygro-Thermometer Model SDL500, serial no. Z335535). After sampling, the chamber-like room and the coughing machine were cleaned with sodium hypochlorite wipes and 70% ethanol.

Virus sample analysis

Viability of the biosampler liquid media was assessed using a plaque assay. The sample was concentrated from ~20 mL to <1,000 μL using an Amicon Ultra 15 centrifugal filter (Cork, Ireland), which was spun in the centrifuge at 4,000 rotations per minute (RPM) in 1-minute intervals. The concentrated sample volume was measured and recorded. A serial dilution was then conducted using 100 μL of the concentrated sample and 900 μL of MS2 broth (Appendix A). Petri dishes (Fisher Scientific, 100 mm \times 15 mm, Polystyrene, MA) for the plaque assay were prepared using 10 mL bottom agar (Appendix A), which was left to rest for 30 minutes before any overlay was added. Top agar was prepared and placed in aliquots in 3-mL tubes (FalconBrand, 14-mL polystyrene round bottom tubes, Mexico), which were then incubated for 20 minutes at 45°C. After the incubation period, 10 μL liquid *Escherichia coli* (10^7 PFU/mL, ATCC15597) and 100 μL serial dilution sample were added to the 3 mL of top agar, which was mixed and poured over the solidified-bottom agar plates. Each serial dilution was plated in triplicate. The plates were left to solidify for 20 minutes and then sealed with a thermoplastic film (Parafilm wrapping film, Bemis, RPI, WI). The sealed plates were placed upside down in a 37°C incubator for 14–24 hours. After the incubation period, the MS2 plaques formed. The serial dilution plate that produced between 20 and 250 plaques were then counted. The arithmetic mean was calculated using the triplicate sample.

Viruses were quantified by counting plaque-forming units (PFU) per unit volume (eg, PFU/mL).^{10,11} Viable virus was the primary concern because it is needed to infect other individuals.¹²

In the case of nosocomial infection, a patient must shed viable virus for the HCP to be infected.¹² The viable virus concentration was calculated from the PFU. The plaque count and dilution factor were used to calculate the plaque concentration (Eq. 1). The plaque concentration, concentrated volume, and air volume sampled were then used to determine the airborne concentration of MS2 (Eq. 2).

$$\text{Plaque Concentration (PFU)} = \frac{(\text{Average plaque forming units})}{(\text{Total volume plated})} \quad (1)$$

$$\text{Sample Concentration (PFU/m}^3\text{)} = \frac{\text{Plaque concentration (PFU)}}{\text{Volume of air sampled (m}^3\text{)}} \quad (2)$$

After all 18 trials were completed, the normality of the samples was tested using the Shapiro-Wilks test. A box plot showing the mean, median, outliers, minimum, and maximum was computed across the 18 trials for the inside and outside of the face shield. A paired *t* test was performed to compare the 2 groups, with a critical *P* set to 0.05.

Particle sample analysis

The OPCs were connected to 2 computers operating the OPC Software (Spectrometer, GRIMM Control, 1179, V1-0-6, 25-10-2018) where particle data were recorded, stored, and exported to Microsoft Excel (Microsoft Office Professional Plus 2016, version 16.0.5278.1000, Redmond, WA) for analysis. The inside and outside particle concentration data for all the trials were compiled for each channel size. The Shapiro-Wilk test was used to assess for normality, after the data were log transformed. Any censored data (data that were recorded as zero) were assigned a value below the limit of detection (LOD, 5 particles/m³).¹³ All OPC zero values were replaced with log (2.5) or 0.39794 (count/m³). Descriptive statistics were calculated using box plots to identify the median of the first quartile, the median of third quartile, standard deviations, and outliers for each experimental condition. For each variable, outliers were identified as values greater than 1.5 interquartile range (IQR) of the third quartile or less than 1.5 IQR of the first quartile. Paired *t* tests were also performed to examine the differences in number particle concentration between the inside and outside of the face shield. The paired *t* test was conducted using an unadjusted α of .05 as well as an α that was adjusted for false discovery rate using the Benjamini Hochberg procedure.¹⁴

Results

Virus concentration

The concentration of the MS2 aerosolized into the coughing machine was 1×10^9 PFU/m³. This concentration was verified after the aerosolization coughing event as well to ensure that it was the same. Air samples were “blank corrected” throughout the experiment. The calculated airborne concentration on the inside of the face shield, near the breathing zone of the HCP, had an arithmetic mean over the 18 trials of $3.54\text{E}+07$ PFU/m³ (SD, $4.20\text{E}+07$) (Table 1). The mean viable virus airborne concentration on the outside of the face shield was $1.15\text{E}+08$ PFU/m³ (SD, $9.76\text{E}+07$). There was a statistically significant reduction ($P < .0006$) in viable virus concentration on the inside of

Table 1. Arithmetic Mean (SD) Concentrations of Viable MS2 Virus on the Inside and the Outside of the Face Shield of the Healthcare Provider During a Simulated Coughing Event

	Outside	Inside
MS2 Concentrations (PFU/m ³)	1.15E+08 (9.76E+07)	3.54E+07 (4.20E+07)
* <i>P</i> < .0006	N = 18	N = 18

the face shield compared to the outside of the face shield, with a relative risk reduction of 69%.

Particle concentration

The number particle concentrations on the inside and outside of the face shield worn by the HCP across all sizes are shown in Figure 2. The particle concentration was significantly lower on the inside of the face shield compared to the outside of the face shield for 12 of the 16 particle sizes measured, using false discovery rate (FDR)-adjusted *P* values ($P < .05$) (Table 2). No significant difference was observed across distances for particle sizes of 0.800–0.943 μm , 0.943–1.112 μm , 1.310–1.545 μm , or 2.982–3.515 μm . Significant differences were observed across distances for particle sizes of 0.253–0.298 μm , 0.298–0.352 μm , 0.352–0.414 μm , 0.414–0.488 μm , 0.488–0.576 μm , 0.576–0.679 μm , 0.679–0.800 μm , 1.112–1.310 μm , 1.545–1.821 μm , 1.821–2.146 μm , 2.146–2.530 μm , and 2.530–2.982 μm .

Discussion

HCP can be exposed to aerosolized SARS-CoV-2 when treating infectious COVID-19 patients; therefore, we need to understand the effectiveness of face shields at protecting HCPs. We analyzed viable virus and particle concentrations across a disposable plastic face shield placed on an HCP during a simulated cough from a patient. We performed a statistical analysis to compare the virus concentrations found across the face shield to analyze the effectiveness of the face shield at reducing virus exposure. These results can be used to inform control measures for protecting HCPs. By performing the sampling experiment in a chamber like room, we were able to limit outside variables, such as drafts, from ventilation and foot traffic to ensure that our results pertained only to the aerosolization event itself.

We observed a statistically significant difference in airborne virus concentration between the inside and outside of the face shield ($P = .0006$). Having a 7-fold reduction in viable virus concentration may not eliminate the risk of infection; however, it may reduce the severity of infection if there is a dose–response relationship; it may reduce the risk of infection by reducing a viral inoculum to below an infectious threshold; or it may increase the effectiveness of medical masks via stepwise reduction in viable aerosolized virus. We observed a statistically significant difference ($P < .05$) in particle number concentrations between the inside and outside of the face shield for most channel sizes. The observed outliers may be due to the variability in particle concentration when operating the coughing machine across experimental trials.

These results align closely with other studies reporting that face shields may not be the most effective method of reducing virus containing aerosols.^{15,16} However, these studies did not analyze

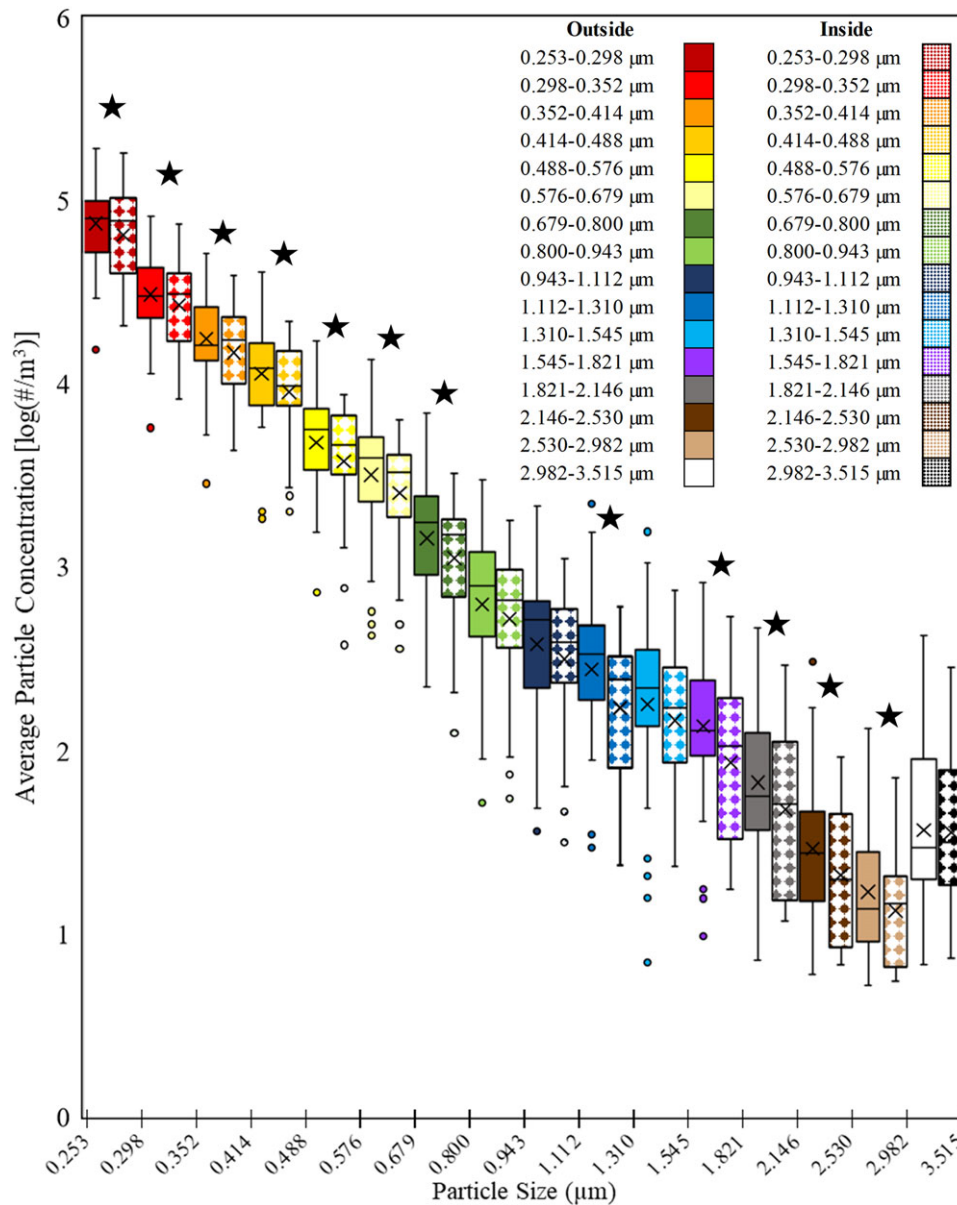


Figure 2. Box plot of average particle concentration comparing the inside and the outside of the face shield placed on HCP exposed to patient cough. The inside of the face shield is solid filled and the outside of the shield is pattern filled. The data are organized by increasing channel size. Statistically significant pairs are marked with stars.

viable airborne virus concentrations, which has implications for disease transmission and effectiveness. Our study demonstrated that virus aerosol-maintained infectivity inside the face shield, representing a risk of disease transmission. Therefore, HCPs may need to wear respiratory protection when treating patients with an active respiratory virus infection presenting with cough.

Face shields protect the HCP from larger aerosolized particles by preventing exposure to the mucous membranes on the face (eg, eyes). However, this study was not designed to assess this exposure. Face shields worn as a barrier protection device will prevent HCP direct contact with splashes and sprays of potentially infectious materials.¹⁷ A face shield also prevents the HCP from touching their eyes, nose, and mouth, which can also lead to indirect transmission and infection.¹⁸ A face shield could still be protective if worn by HCPs when treating potentially infectious patients. A limitation of our study design was that we used a bluff body to

represent the HCP instead of a breathing mannequin due to the unavailability of a simulated breathing machine. However, we chose a sampler that represented a breathing rate similar to humans to minimize this limitation. We expect that if a breathing machine had been implemented it would have driven the results toward the null because more mixing would have occurred. Future experiments could be done with a breathing machine as the HCP to improve upon these findings. In addition, the coughing machine, representing a simulated patient, produced an exhaled cough and did not inhale air. Future studies should generate a coughing machine that can both inhale and exhale.

Another limitation was using a plaque assay to determine the viable aerosolized virus concentration to estimate infection risk. Due to the mechanical nature of the coughing machine, some of the viable virus could be inactivated during aerosolization, simulated coughing and sampling. This error did not likely affect

Table 2. Paired *t* Test Results for Comparing the Inside and Outside of the Face Shield Across Channel Size When the Face Shield is Placed on the Simulated HCP

Channel size (μm)	Location	Mean	Variance	df	t Stat	Unadjusted <i>P</i> value	FDR adjusted <i>P</i> Value
0.253–0.298	Outside	4.875	0.057	28	2.984	<.01*	<.01*
	Inside	4.814	0.063				
0.298–0.352	Outside	4.489	0.061	28	2.444	.021*	.03*
	Inside	4.429	0.064				
0.352–0.414	Outside	4.245	0.070	28	2.800	<.01*	<.01*
	Inside	4.171	0.063				
0.414–0.488	Outside	4.055	0.087	28	3.135	<.01*	<.01*
	Inside	3.955	0.076				
0.488–0.576	Outside	3.679	0.104	28	2.847	<.01*	<.01*
	Inside	3.577	0.110				
0.576–0.679	Outside	3.501	0.135	28	2.957	<.01*	<.01*
	Inside	3.403	0.109				
0.679–0.800	Outside	3.156	0.147	28	2.869	<.01*	<.01*
	Inside	3.048	0.134				
0.800–0.943	Outside	2.794	0.181	28	1.711	.098	.112
	Inside	2.717	0.161				
0.943–1.112	Outside	2.578	0.201	28	1.740	.093	.112
	Inside	2.499	0.180				
1.112–1.310	Outside	2.441	0.198	28	3.356	<.01*	<.01*
	Inside	2.232	0.180				
1.310–1.545	Outside	2.250	0.279	28	1.624	.116	.123
	Inside	2.165	0.182				
1.545–1.821	Outside	2.131	0.210	28	3.233	<.01*	<.01*
	Inside	1.935	0.196				
1.821–2.146	Outside	1.822	0.193	28	2.225	.034*	.045*
	Inside	1.676	0.214				
2.146–2.530	Outside	1.463	0.143	28	2.562	.016*	.029*
	Inside	1.319	0.142				
2.530–2.982	Outside	1.228	0.135	28	2.379	.024*	.036*
	Inside	1.126	0.096				
2.982–3.515	Outside	1.565	0.214	28	0.352	.728	.728
	Inside	1.553	0.185				

Note. HCP, healthcare personnel; FDR, false discovery rate; df, degrees of freedom.

*Statistically significant (<.05).

our experimental observations because we used the same methods to assess both the inside and the outside of the face shield. We also reduced the impact of experimental variability in the plaque assays by running each serial dilution in triplicate and averaging the concentration across trials.

MS2 may not be representative of SARS-CoV-2 virus. MS2 has been used successfully as a surrogate for SARS-CoV-2 as well as norovirus, calicivirus, and poliovirus.^{6,8,9} In addition, MS2 is a positive-sense, single-stranded, RNA phage like SARS-CoV-2.¹⁹ MS2 is smaller than SARS-CoV-2 (23–28 nm vs 100 nm), but the particles that transmit through coughing are likely much larger because they occur in conglomerations of liquid particles.¹⁹ MS2 is also nonenveloped whereas SARS-CoV-2 is enveloped, meaning that it may have different biophysical properties such as susceptibility to dehydration. Future experiments should confirm the results

of this study using an enveloped surrogate like Phi 6 or with SARS-CoV-2.

Also, we also did not correct for background aerosol concentration in the chamber, so some of the particles measured might have been from room background aerosol. This limitation did not affect the results of our experiment because we measured live virus concentrations across the face shield rather than solely relying on airborne particle concentrations.

In conclusion, a face shield worn on an HCP was effective at reducing exposure to virus containing aerosol from simulated coughing. Future studies should be conducted to evaluate other scenarios (eg, sneezing, tidal breathing etc) and PPE ensembles (eg, N95 and face shield combination) using simulated breathing machine. In addition, future studies quantifying the benefit that face shields provide for eye protection are warranted.

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Competing interests. All authors report no Competing interests relevant to this article.

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Appendix A

MS2 Broth Solution Recipe

Ingredient	Broth	Bottom Agar	Top Agar
Tryptone (g)	5	5	5
Yeast Extract (g)	0.5	0.5	0.5
NaCl (g)	4	4	4
Agar (g)	0	7.5	2.5
Supplement (mL)	25	25	25
DI Water (mL)	475	475	475

Makes 500mL. All solutions were autoclaved without added supplement at 121°C for 20 minutes then placed in a water bath (55°C) to cool. The supplement was added and mixed once the solution cooled.

Solution Recipe of Supplement

Ingredient	Supplement
Glucose (g)	10
CaCl ₂ (g)	2.934
Thiamine [20]	100
DI Water (mL)	500

Makes 500 mL. Filter solution with 0.22-µm pore filter prior to use.