SAND2020-5621R

Sandia National Laboratories

Self-Disinfecting Polymeric Coatings

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June 2020

LDRD LABORATORY DIRECTED RESEARCH & DEVELOPMENT
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EXECUTIVE SUMMARY

A novel derivative of a previously-published polymeric material has been synthesized and developed into an easily-sprayable coating. Surface characterization of coatings confirm correct elemental presence, and viral assays reveal quantitative elimination of MS2 bacteriophage and Phi6 bacteriophage, surrogates used for SARS-CoV-2, in as little as 5 minutes upon contact. Furthermore, an N95 mask was dip-coated in the polymer solution and analyzed through microscopy and filtration efficacy testing. Though coating was successful, electrostatic interactions between mask layers and polymer reduced filtration efficacy significantly. As such, we expect the current results of this work to be applicable on non-respiratory PPE and on solid substrates of commonly-touched surfaces for rapid self-decontamination.

ACKNOWLEDGEMENTS

Supported by the Laboratory Directed Research and Development program at Sandia National Laboratories, a multimission laboratory managed and operated by National Technology and Engineering Solutions of Sandia LLC, a wholly owned subsidiary of Honeywell International Inc. for the U.S. Department of Energy's National Nuclear Security Administration under contract DE-NA0003525.

ACRONYMS AND DEFINITIONS

Abbreviation	Definition
CFR	Code of Federal Regulations
CPC	Condensation Particle Counter
DMA	Dynamic Mechanical Analysis
EDS	Energy-Dispersive X-Ray Spectroscopy
FPT	Filter Penetration Testbed
IPA	Isopropyl Alcohol
LB	Luria Broth
MEK	Methyl Ethyl Ketone
MIT	Massachusetts Institute of Technology
MTBE	Methyl Tert-Butyl Ether
N-DMPEI	N,N-Dodecyl-Methyl-Polyethyleneimine
NEB	New England Biolabs
NFPA	National Fire Protection Association
N-HMPEI	N,N-Hexyl-Methyl-Polyethyleneimine
NIOSH	National Institute for Occupational Safety and Health
PEI	Polyethyleneimine
PE	Polyethylene
PET	Polyethylene Terephthalate
PFU	Plaque-Forming Unit
PP	Polypropylene
PPE	Personal Protective Equipment
RNA	Ribonucleic Acid
SARS-CoV-2	Severe Acute Respiratory Syndrome Coronavirus 2
SEM	Scanning Electron Microscopy
SNL	Sandia National Laboratories

Background

Sprayable polymeric coatings that have self-disinfecting properties towards viruses, bacteria, and other microbes are needed to provide facile decontamination of commonly-touched surfaces. Such materials would ideally require no input or stimuli from relevant personnel; the chemical structure of the polymer would act as a solid-state soap to destroy microbes by disrupting membranes or envelopes surrounding sensitive biological material. Polymers with these properties were prepared by Park et al at MIT back in 2006. Their work showed quantitative efficacy in killing both *S. aureus* and *E. coli* bacterial lines using branched 750 kDa *N*-alkyl-polyethyleneimines (N-alkyl PEIs).[1] Furthermore, they proved the mechanism of destruction was through the rupturing of the bacterial membranes. They expanded on their work later that year by testing efficacy of similar materials towards the WSN strain of the influenza virus.[2] The results of their work proved that 100 % vircudal activity is achieved in as little as 5 minutes (Figure 1).

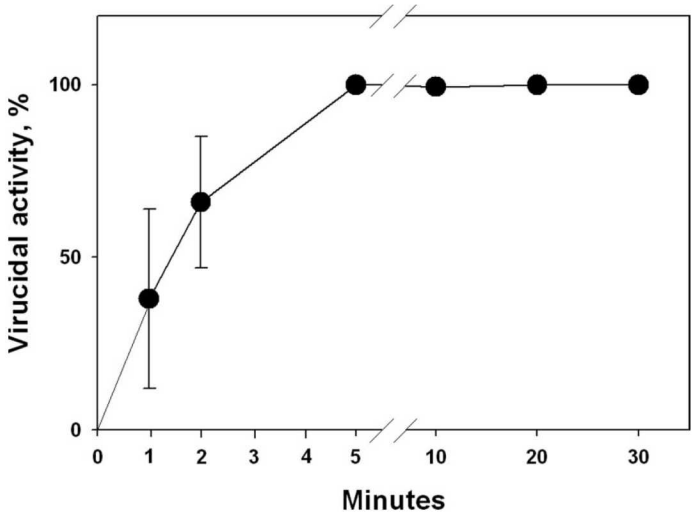


Figure 1. The time course of inactivation of influenza virus (WSN strain) by a glass slide painted with a linear 217 kDa *N*,*N*-dodecyl-methyl PEI at room temperature.[2]

This COVID-19 project seeked to utilize these proven materials in an easily-sprayable coating. Commercial materials have changed over the past 14 years, thus branched 270 kDa PEI was used as the starting material rather than the 750 kDa branched PEI or 217 kDa linear derivatives seen in MIT's work. Coating application onto aluminum substrates and immersion of N95 respirators into polymer solutions were performed to evaluate extent of use of these methods and materials. Other virus surrogates were also used to test the virucidal efficacy of the synthesized polymer materials, specifically MS2 bacteriophage and Phi 6 bacteriophage (Table 1).

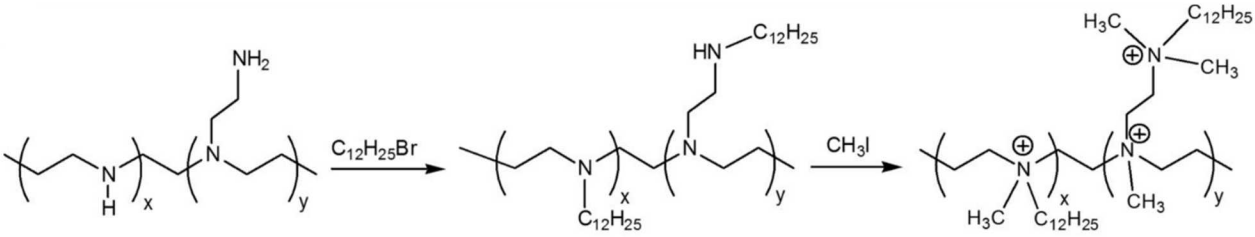
Table 1. Structural and property comparisons of phi6, MS2, and SARS-CoV-2.

Phi 6	**MS2**	SARS-CoV-2
Seven structural proteins	One structural protein	Four structural proteins
Lipid envelope (virion is 20% lipid)	No lipid	Lipid envelope
Double strand + RNA	Single strand + RNA	Single strand + RNA
13,379 nucleotides	3,569 nucleotides	30,000 nucleotides
Surface spikes	No spikes	Surface spikes
No glycoproteins	No glycoproteins	
Spherical 86 nm	Spherical 24-26 nm	Spherical 50-200nm
Icosahedral nucleocapsid, 43 nm	Icosahedral nucleocapsid, 24-26 nm	
Stable at physiological and mild alkaline pH	Stable at physiological and mild alkaline pH	
Prokaryotic host Pseudomonas syringae	Prokaryotic host, e.g. Escherichia coli	

Synthesis of *N,N*-Hexyl-Methyl Polyethyleneimine (*N*-HMPEI) and *N,N*-Dodecyl-Methyl Polyethyleneimine (*N*-DMPEI)

All materials were of ACS reagent grade or greater and used as received from Sigma-Aldrich. Branched polyethyleneimine (PEI) was used with an average Mw ~ 270 kDa. Scheme 1 shows the structural changes during the two-step synthesis of the active material.

Scheme 1. Two-step synthetic prep for N-alkyl PEIs[1]



This procedure was directly followed according to the literature, only scaled up.[1] A solution of 16.5 g of PEI and 63 g of K_2CO_3 in 210 mL of *tert*-amyl alcohol and 53 or 90 mL of 1-bromohexane and 1-bromododecane, respectively, were mixed and stirred at 95 °C for 24 and 96 hours. After removing the solids by filtration under reduced pressure, 45 mL of iodomethane was added, followed by stirring at 60 °C for 24 h in a sealed bottle. The resultant solution was added to an excess of *n*-hexane for *N*-HMPEI and acetone for *N*-DMPEI and the precipitate was recovered by filtration under reduced pressure, washed with an excess of *n*-hexane and acetone, respectively, and dried at room temperature under vacuum overnight. Product was ground in a mortar and pestle to yield a faint yellow powder.

Polymer Dissolution Studies

An assortment of volatile solvents were purchased to study the solubility of the two polymer derivatives for coating purposes. The molecular structure of the polymers make for interesting and complex solubilities due to the presence of both ionic character and long aliphatic chains. All solvents tested had NFPA health ratings of 1; no solvents with a 2 or higher were used due to the expected use of the materials. Dissolution testing was performed at 1 weight % for all samples. Table 2 illustrates the results of the studies.

Table 2. Solubility Results of 1 wt % PEI Derivatives.

Solubility Results		
Solvent	***N*-HMPEI**	***N*-DMPEI**
Water	No	No
Methanol	Yes	No
Ethanol	Yes	No
2-Propanol (IPA)	Yes	No
1-Propanol	Yes	No
2-Butanol	Yes	Yes
2-Butanone (MEK)	No	Yes
Methyl *tert*-butyl Ether (MTBE)	No	Yes
n-Heptane	No	No

A clear distinction in solubility is seen between the two derivatives due exclusively to the alkyl chain length difference. The shorter hexyl chains are expected to allow interactions to occur between the ionic components of the polymer while the longer dodecyl chains provide steric hindrance to those groups. 2-butanol was the only solvent that solubilized both polymers, and neither polymer was soluble in water, which is a key component for the coating application on commonly-touched surfaces.

SEM/EDS (Scanning-Electron Microscopy/Energy Dispersive X-Ray Spectroscopy)

To determine the elemental makeup of the polymer materials, SEM/EDS was performed (Figures 2 and 3). Aluminum coupons with dimensions of one square inch were sprayed five times with 1 wt% polymer solution of *N*-HMPEI in methanol or 1 wt% *N*-DMPEI in MTBE using a fine mist sprayer. Coatings were allowed to dry in a fume hood at room temperature. All images were obtained using a Zeiss GeminiSEM 500 Variable Pressure Field Emission Scanning Electron Microscope equipped with a Bruker XFlash 6|60 SDD Energy Dispersive Spectrometer. Bruker Esprit analysis software was used for EDS collected. For the EDS maps- 10 kV accelerating voltage was used in order to detect the iodine with the 60-um aperture and high volt age option selected. For imaging, 5 kV accelerating voltage was used with the 20-um aperture selected. Au-Pd was applied to provide a conductive coating.

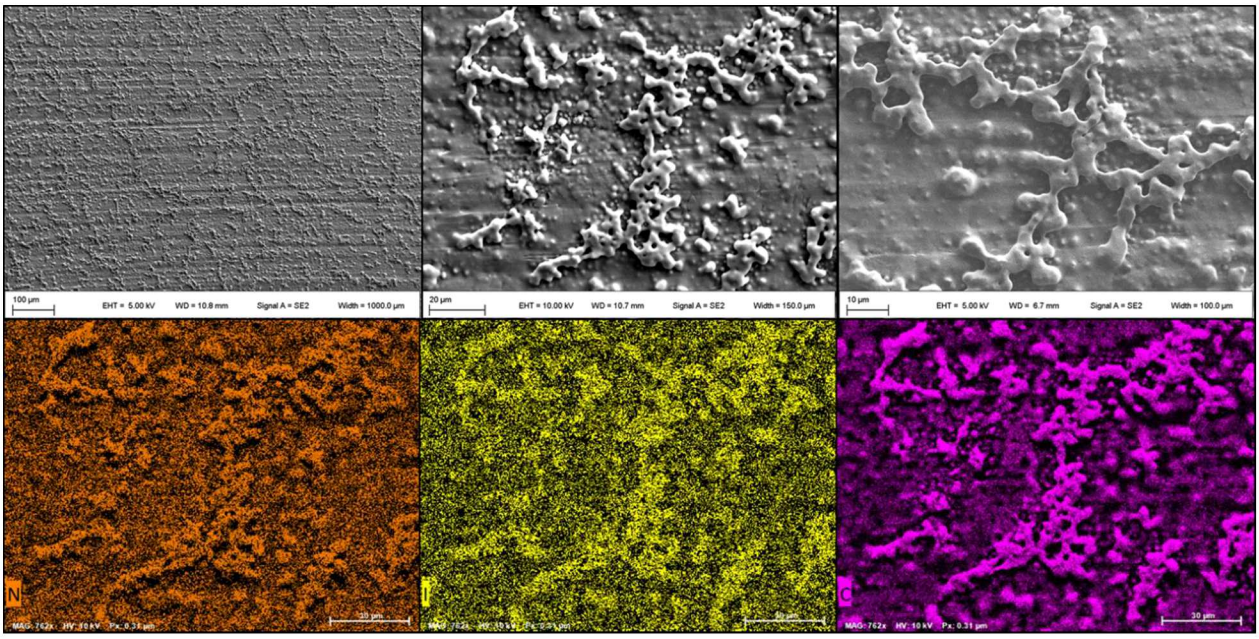


Figure 2. SEM (top) and EDS (bottom) of *N*-HMPEI on an aluminum substrate. Nitrogen (orange), iodine, (yellow), and carbon (purple) EDS images taken from middle picture.

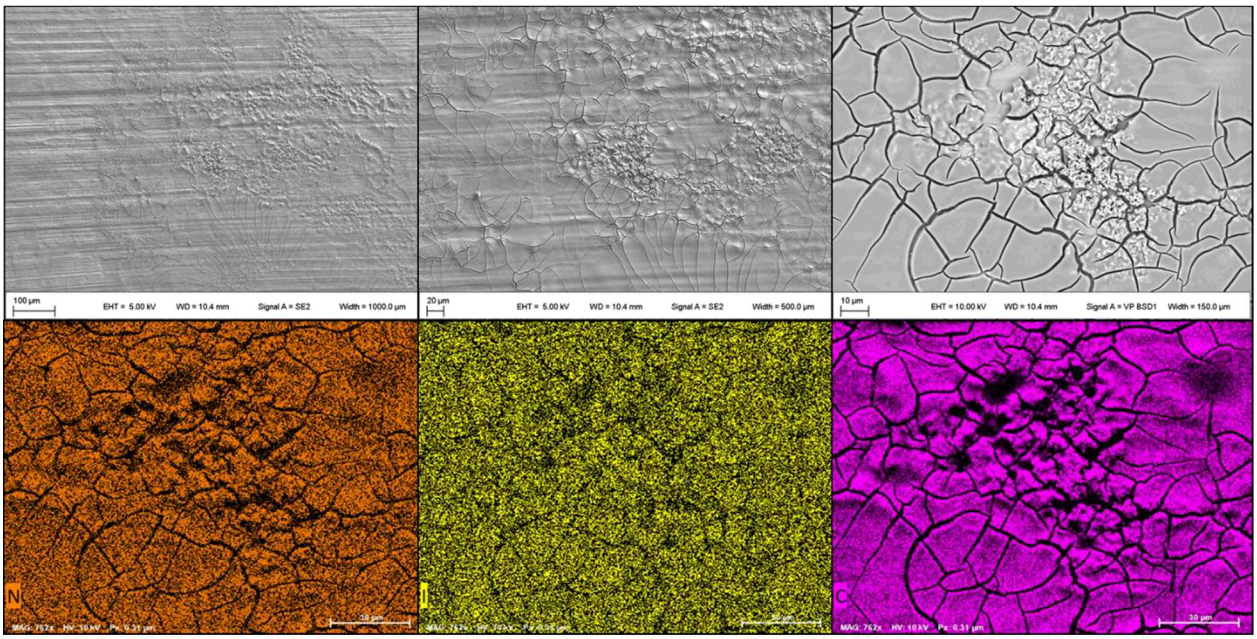


Figure 3. SEM (top) and EDS (bottom) of *N*-DMPEI on an aluminum substrate. Nitrogen (orange), iodine, (yellow), and carbon (purple) EDS images taken from right picture.

SEM images of the two different solutions indicate good surface coverage throughout the samples. The *N*-HMPEI sample, which was dissolved in methanol, shows 100% coverage with no cracking of the surface. This may imply the evaporation rate of methanol is ideal to achieve the appropriate coating surface. The *N*-DMPEI sample, dissolved in MTBE, showed nanoscale-thin cracks throughout the coating. Of all the solvents used in the solubility study MTBE had the most volatility. The rapid rate of evaporation may have led to stress fracturing of the polymer coating surface. To evaluate this, a 1 wt% solution of both polymers were made in 2-butanol, the only solvent where both materials show equivalent solubility. Compared to methanol and MTBE, which have vapor pressures of ~ 13 and 33 kPa, respectively, 2-butanol has a vapor pressure of ~ 2 kPa. The lower vapor pressure, and thus evaporation rate, were expected to yield surface coatings with no cracks. Surprisingly, cracks were also observed from these samples (appendix).

EDS mapping of the sample surfaces revealed all elements expected to prove polymer coating was applied uniformly. Alkyl-PEIs are composed exclusively of nitrogen, carbon, and hydrogen, with the counter anions being iodine. Excluding hydrogen, which cannot be detected by EDS, all elements were visible and clearly dispersed over the entire surface.

Optical Imaging

Optical images of polymer coatings on 3M 8511 N95 respirator layers were obtained via optical imaging using a Keyence VHX6000 and a 250-2500x objective. Images were taken of a control mask sample and the sample dipped in the 1 wt% *N*-DMPEI in MTBE solution.

Optical images of respirator samples (Figure 4) show a complex network of interweaving fibers that have differing morphologies dependent on the layer. The chemical identities of the layers are polyethylene terephthalate (PET), PP, PP/PE, PP/PE, and PP, respectively. Bonding of polymers seems to differ based on the type of layer. Layers 1 and 2 have similar coatings that are visibly dispersed around the fibers themselves. It is expected that, even though the yellow polymer is not fully visible on the fibers, a thin film exists on all surfaces. Layer 5 tends to show more polymer deposit particles than coating on the fibers. Finally, layers 3 and 4 are the PP electret components. Based on the electrostatic testing performed on the respirator layers (next section), the electret has a net negative charge associated with it. The polymers, being positively charged, electrostatically bond to these fibers, which is why their coverage is the greatest of all. Neutralization of the electrostatic charge occurs, however, due to this surface coverage (discussed later).

Optical images also deduced that each layer of the respirator has two separate layers associated with it, an outer layer (closest to environment) and an inner layer (closest to face). Based on the images obtained, all inner and outer layers were coated except for the inner of 3 and the outer of 4 (Figure 5). As these are the electret layers and both uncoated layers are in the same vicinity, it is expected that outer layer 3 and inner layer 4 acted as electrostatic filters towards the polymer materials, bonding all available material that was exposed in the 10 second dip of the mask in the solution. Longer dip times and/or more concentrated solutions would most likely aid in full coating application of respirator layers.

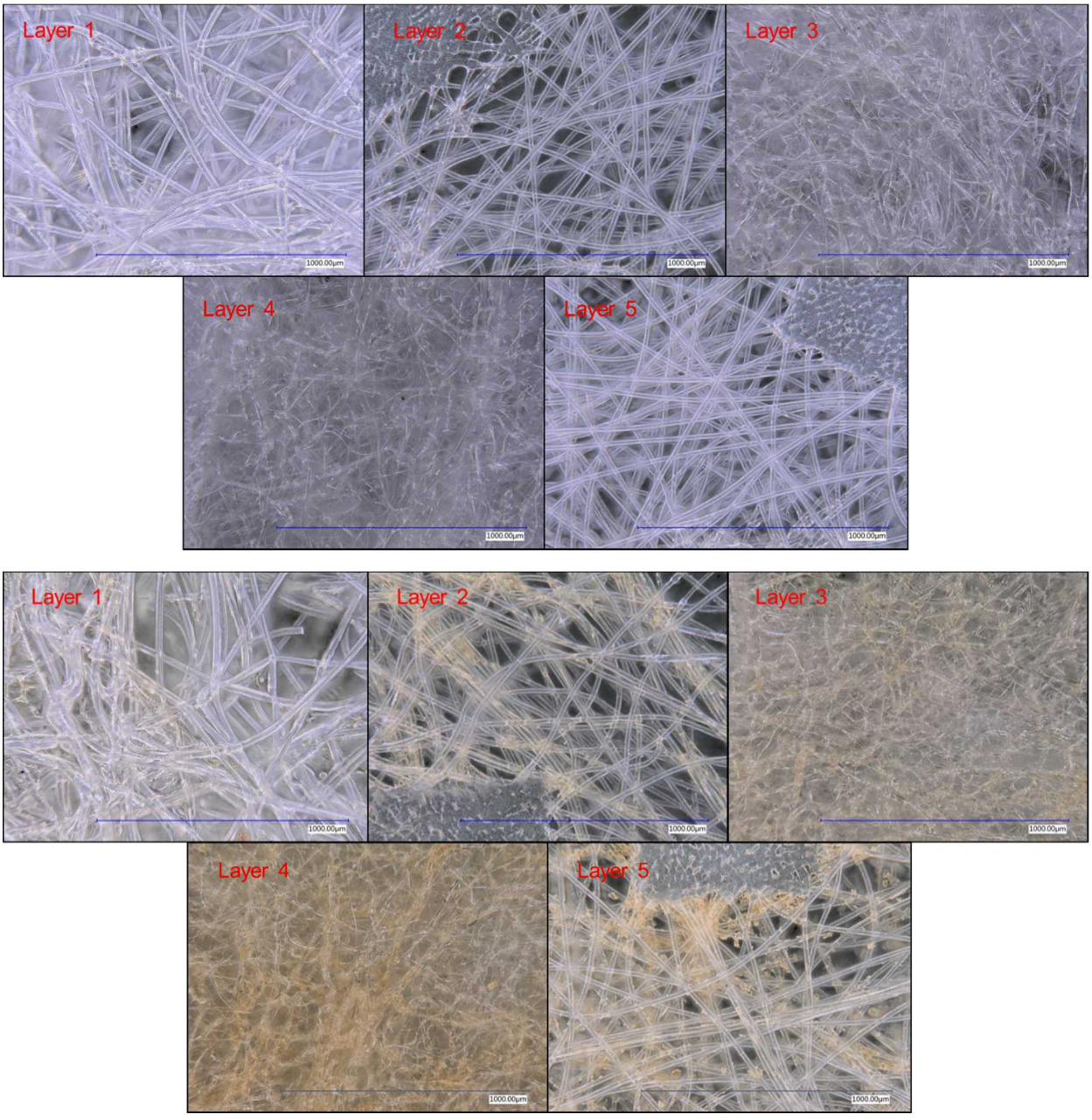


Figure 4. Optical images of uncoated respirator layers (top) and respirator layers coated with 1 wt% *N*-DMPEI in MTBE. All images use a 250x objective and all scale bars = 1000 μm.

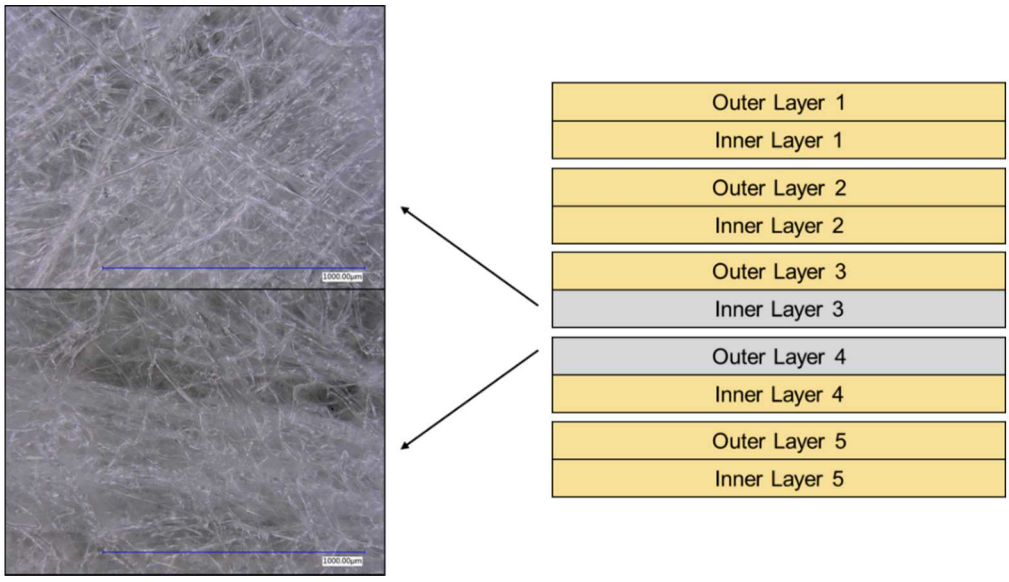


Figure 5. Respirator fabric layers with polymer coatings. Two sublayers show no coating applied. Scale bars = 1000 µm.

Electrostatic Testing

All measurements were made with a Trek 821HH contact electrostatic probe. The probe was grounded to earth ground between each measurement. All voltages are measured versus earth ground. A minimum of five measurements were taken on each layer (with a few exceptions). Solvents used for polymer dissolution were also tested separately to determine their effects on the electret material (10 second immersions). IPA sample was soaked for one hour and was used as a positive fail control sample. Figure 6 provides the graphical summary of the results obtained from the testing.

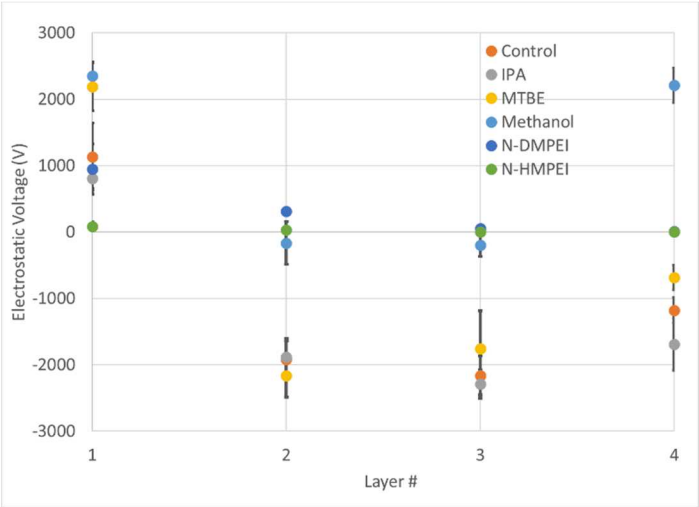


Figure 6. Electrostatic testing results of material layers seen in a 3M 8511 N95 respirator. Polymer solutions (green and dark blue) zeroed out much of the charge of all the layers.

The solvents chosen for dissolving the polymers were based on 1) solvents predetermined that solubilize each polymer derivative and 2) the absolute value difference in Hansen solubility parameters (δ) of the solvent in question and polypropylene (PP), the electret material in the mask. A large solubility parameter difference was expected to cause less of an effect on the mask material due to polarity differences. The δ values, respectively, for PP, methanol, and MTBE are 18.8, 29.7, and 7.4. The absolute value difference between the PP and the solvents is thus 10.9 and 11.4, respectively, which basically should eliminate the solvent difference between the two samples. A one hour immersion in IPA (δ difference = 5.7), which was used as the positive control, surprisingly shows only a modest effect on the charge of the respirator materials. MTBE had a similar effect, which was expected. Methanol, however, had an unexpectedly larger effect. The polymer materials themselves, being ionic, had a significant effect on the respirator layers and eliminated most of the charge present. Steric interactions could be playing a small role here between the N-HMPEI and N-DMPEI since the N-DMPEI had less of an effect comparatively.

Filtration Efficiency

A filter efficiency study was conducted on the Filter Penetration Testbed (FPT) located at SNL. The testbed was designed to closely follow NIOSH guidance and the Code of Federal Regulations (CFR) set forth in 42 CFR, Part 84, Subpart K, §84.181. The FPT size selects for a monodispersed sodium chloride, NaCl, aerosol by utilizing a TSI 8020 Electrostatic Classifier and TSI 3081L Differential Mobility Analyzer (DMA) upstream from a 47 mm stainless filter housing. NIOSH guidance specifically mentions a monodispersed aerosol with particle size median diameter of 0.075 ± 0.020 μm with a geometric standard deviation not exceeding 1.86 that has been neutralized to the Boltzmann equilibrium state, which the FPT system met. However, it is important to note that the FDA only requires tests to be conducted at a monodispersed particle size of 0.3 μm, but specifically mentions 42 CFR, Part 84 for respirator qualification. Additionally, the filter housing diameter used by the FPT is smaller than the mounts used by TSI 8130 (120 mm). Therefore, flow rates were adjusted according to equivalent filter face velocities (17.337 cm/s), so geometry of the filter housing was mitigated. Finally, in order to calculate filter efficiency, concentration was measured upstream and downstream of the filter material tested by a TSI 8022 Condensation Particle Counter (CPC). Five tests upstream and downstream were conducted with one minute intervals, as is mentioned the NIOSH guidance. Efficiency tests are reported below (Table 3) for several 3M 8511 facial mask samples.

Filtration efficiency of polymer-coated samples dropped significantly, which was expected due to the electrostatic testing results. These results are entirely due to the compromised electrostatic nature of the electret layers. Since this property is what filters the smaller particles the efficiency loss is expected. The solvents also had an effect on the electret properties as well, though not to the extent of the polycationic polymers.

Table 3. Filtration efficacy results of polymer-coated N95 masks and respective solvents. * = 10 second immersion; ** = 1 hour immersion

Manufacture	Model	Particle Size	Sample	Filter Velocity (cm/s)	Efficiency (%)	Stdev (%)
3M	8511	75 nm	Control	17.337	99.19	0.13
			N-DMPEI*	17.337	44.99	4.88
			N-HMPEI*	17.337	40.33	1.71
			Methanol*	17.337	82.66	2.10
			MTBE*	17.337	69.23	1.13
			IPA**	17.337	59.30	1.54

Plaque Assays with MS2 Bacteriophage

Media used for experiments included 1% LB agar plates, Lambda diluent phage buffer, and 0.7% LB top agar supplemented with 10 mM Mg^{2+} and 5 mM Ca^{2+}. Biological samples prepared were MS2 phage incubated in pre-treated plates and NEB Turbo II E. coli.

On day 1, NEB Turbo II E. coli was incubated overnight at 37 °C shaking at 200 rpm. On day 2, 100 µL of MS2 Phage (1.3 x 10^{10} PFU/mL) was pipetted into wells of plate (see diagram in appendix for plate set up, one well plate used per chemical treatment of Corning Costar® 24-well plates, cat# 3738). Samples were incubated at 25 °C with 60% relative humidity for at 5, 15, and 30 minute timepoints. A 45 minute timepoint was also taken on the second run of the experiment. Phage was pipetted out of wells of plate at desired timepoint and placed in an epi tube. 12 well plates were returned to incubator and repeated until all timepoint samples were complete. Phage samples were serially diluted to 10^9. Top agar 0.7% was kept warm in a 55 °C bead bath. Next, 4 mL of top agar was portioned out into conical tubes for each plate and kept in the 55 °C bead bath. One plate per timepoint was needed; 100 µL O/N NEB Turbo II E. coli was combined with 4 mL of top agar and the mixture quickly poured over LB agar plates. Plates were swirled so that the whole surface of the plate was covered with top agar. After solidification, 10 µL of each dilution was spotted onto top agar. Spots were allowed to dry and plates were inverted and incubated at 37 °C overnight. Spots formed small 1-2 mm clearings, plaques, in the bacterial lawn,

and counted at each time point for all the samples tested. Plaque forming units (PFU/mL) was calculated by the following equation: PFU/mL= # plaques counted / (vol spotted in mL)(dilution factor). Virucidal activity was calculated by the following equation: 100 – [(Sample count) / (Control count) x 100 %]. Tables 4 and 5 provide the quantitative PFU/mL plaque counts for the samples and Figures 7 and 8 graphically represent the virucial activities versus time.

Table 4. Experiment 1 of MS2 Plaque Assay Results and Virucidal Activity

MS2 Exp.1 5/1/20	*N*-DMPEI (PFU/mL)	*N*-HMPEI (PFU/mL)	Benzalkonium Chloride (PFU/mL)	Control (PFU/mL)	Virucidal Activity *N*-DMPEI	Virucidal Activity *N*-HMPEI
5 min	$2.0x10^8$	$6.0x10^6$	No timepoint taken	No timepoint taken	92.3 %	99.8 %
15 min	$5.0x10^8$	$4.0x10^7$	No timepoint taken	No timepoint taken	80.8 %	98.5 %
30 min	$1.9x10^9$	$2.0x10^6$	0.00	$2.60x10^9$	26.9 %	99.9 %
45 min	N/A	N/A	No timepoint taken	No timepoint taken	N/A	N/A

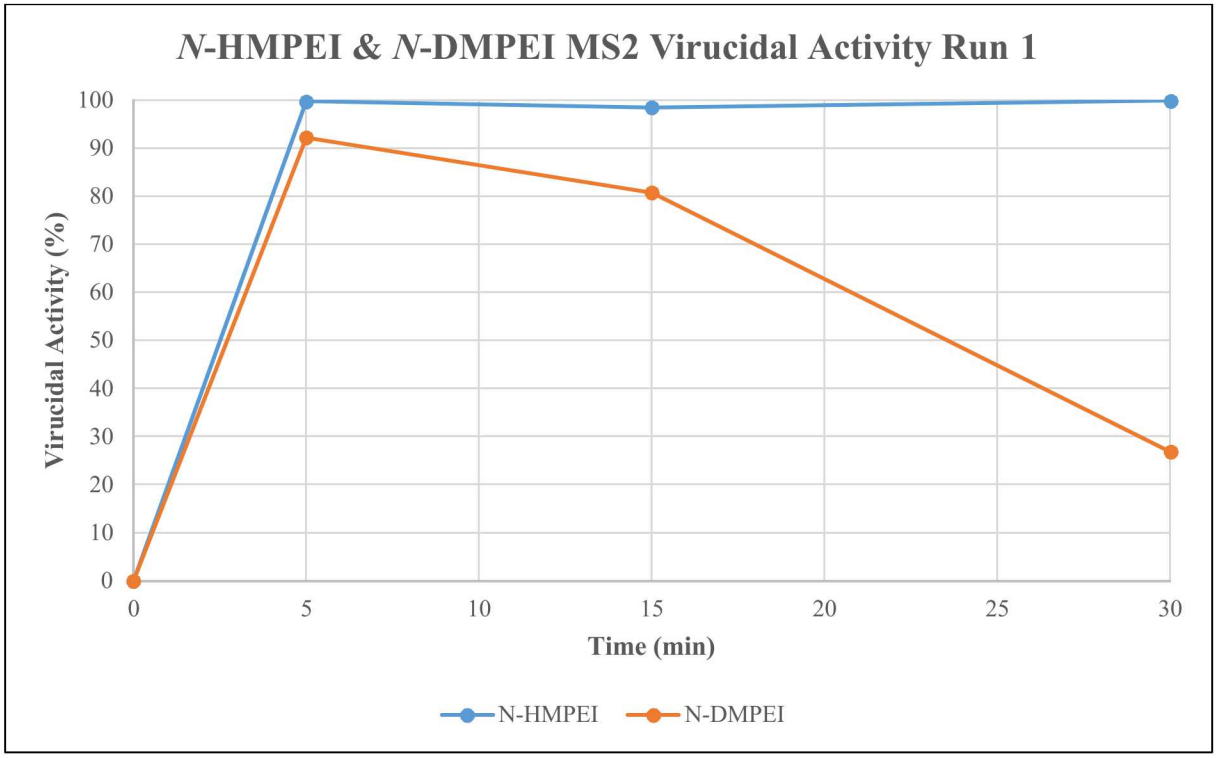


Figure 7. First trial of the time course of inactivation of MS2 phage by a glass slide sprayed with branched 270 kDa *N*,*N*-alkyl-methyl PEI at room temperature.

Table 5. Experiment 2 of MS2 Plaque Assay Results and Virucidal Activity

MS2 Exp. 2 5/7/20	*N*-DMPEI (PFU/mL)	*N*-HMPEI (PFU/mL)	Benzalkonium Chloride (PFU/mL)	Control (PFU/mL)	Virucidal Activity *N*-DMPEI	Virucidal Activity *N*-HMPEI
5 min	$8.0x10^6$	$5.0x10^5$	TNTC-cleared bacterial lawn	$8.0x10^7$	90.0 %	99.4 %
15 min	$6.0x10^6$	$5.0x10^4$	No plaques	$7.0x10^7$	91.4 %	99.9 %
30 min	$6.5x10^7$	$4.0x10^4$	No plaques	$2.0x10^7$	No inhibition	99.8 %
45 min	$4.0x10^6$	$3.0x10^4$	No plaques	$4.0x10^7$	90.0%	99.9 %

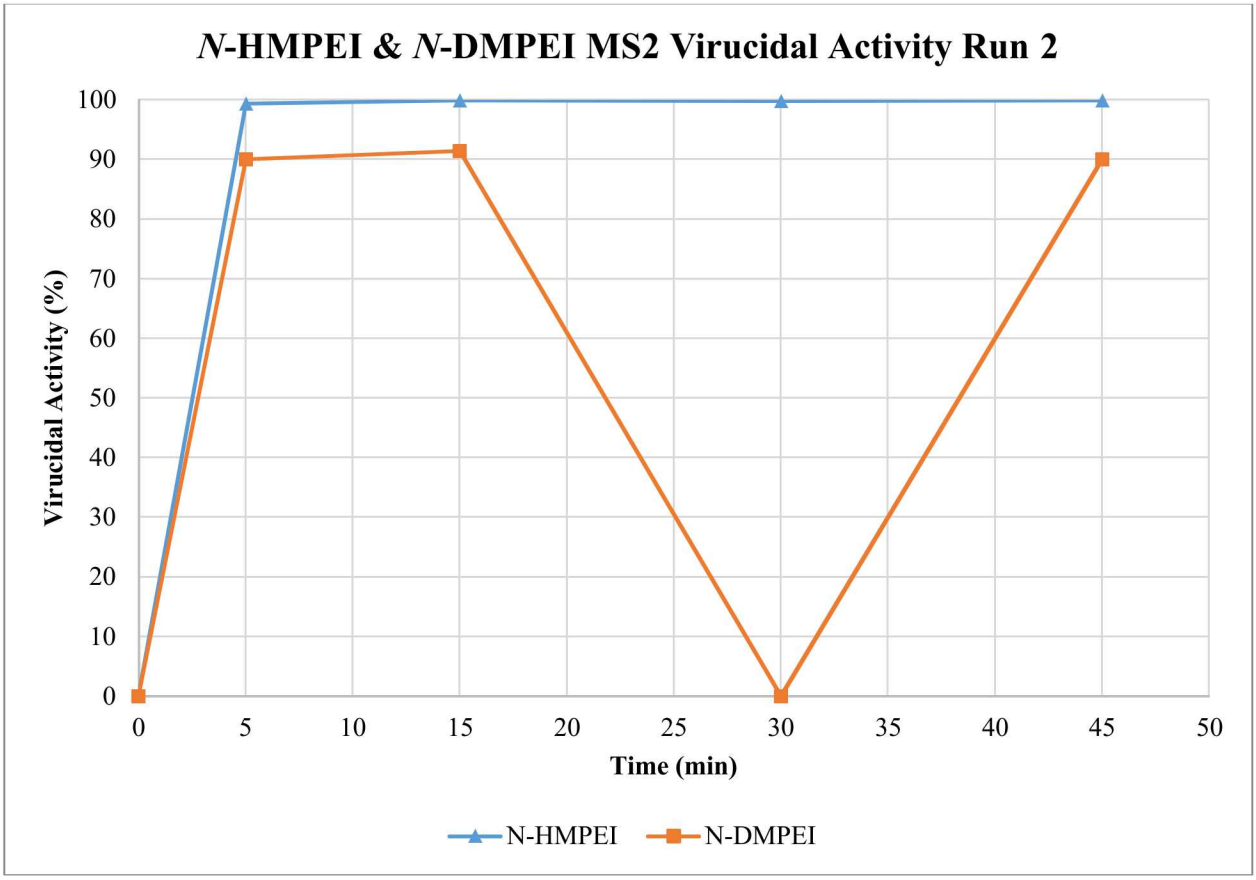


Figure 8. Second trial of the time course of inactivation of MS2 phage by a 24 well plate sprayed with branched 270 kDa *N*,*N*-alkyl-methyl PEI at room temperature.

Overall, lower plaque forming units (PFU/mL) values were seen for phage samples exposed with *N*-HMPEI than those treated with *N*-DMPEI. *N*-DMPEI clearly has a random distribution of data that yields no clear trend. *N*-HMPEI, however, has consistent and efficient virucidal activity values up to 99.9% in a very short amount of time. PFU/mL values for the positive control, benzalkonium chloride, were not obtained as the reagent virtually eliminated the majority of the phage sample. The negative control contained the PFU/mL for phage with no chemical treatment.

Plaque Assays with Phi6 Bacteriophage

Media used for experiments included 1% LB agar plates, Lambda diluent phage buffer, and 0.7% LB top agar. Biological samples prepared were phi 6 phage from UMICH and *Pseudomonas syringae*.

On day 1, *Pseudomonas syringae* was cultured and incubated at 26 °C shaking at 100 rpm. This slow grower takes 48 hours for culture to come up to ideal concentration of cells. On day 3, 100 µL of Phi6 (1.0×10^7 PFU/mL) was incubated on four individual 24 well plates (Corning Costar® 24-well plates, cat# 3738); 3 polymer samples and one control plate all at 5, 15, 30, and 45 min. Phage was serially diluted in phage buffer after each incubation time point. Next, 100 µL of 2 day growth of *Ps syringae* was plated and mixed in 4 mL of 0.7% top agar and poured over LB agar plates. Serial dilution of phage was spot tittered and allowed to incubate at 25 °C for two days. Number of plaques were counted to calculate phage titer. PFU/mL was calculated by the following equation: PFU/mL= # plaques counted / (vol spotted in mL)(dilution factor). Virucidal activity was calculated by the following equation: 100 – [(Sample count) / (Control count) x 100 %]. Table 6 provides the quantitative PFU/mL plaque counts for the samples and Figure 9 graphically represent the virucial activities versus time.

Table 6. Phi6 Plaque Assay Results and Virucidal Activity

Phi6 Exp 5/24/20	*N*-DMPEI (PFU/mL)	*N*-HMPEI (PFU/mL)	Benzalkonium Chloride (PFU/mL)	Control (PFU/mL)	Virucidal Activity *N*-DMPEI	Virucidal Activity *N*-HMPEI
5 min	1.0×10^6	No plaques	No plaques	2.2×10^6	55.55 %	100 %
15 min	1.3×10^6	No plaques	No plaques	2.2×10^6	40.91 %	100 %
30 min	1.3×10^6	No plaques	No plaques	7.0×10^5	No inhibition	100%
45 min	3.8×10^6	No plaques	No plaques	2.5×10^6	No inhibition	100 %

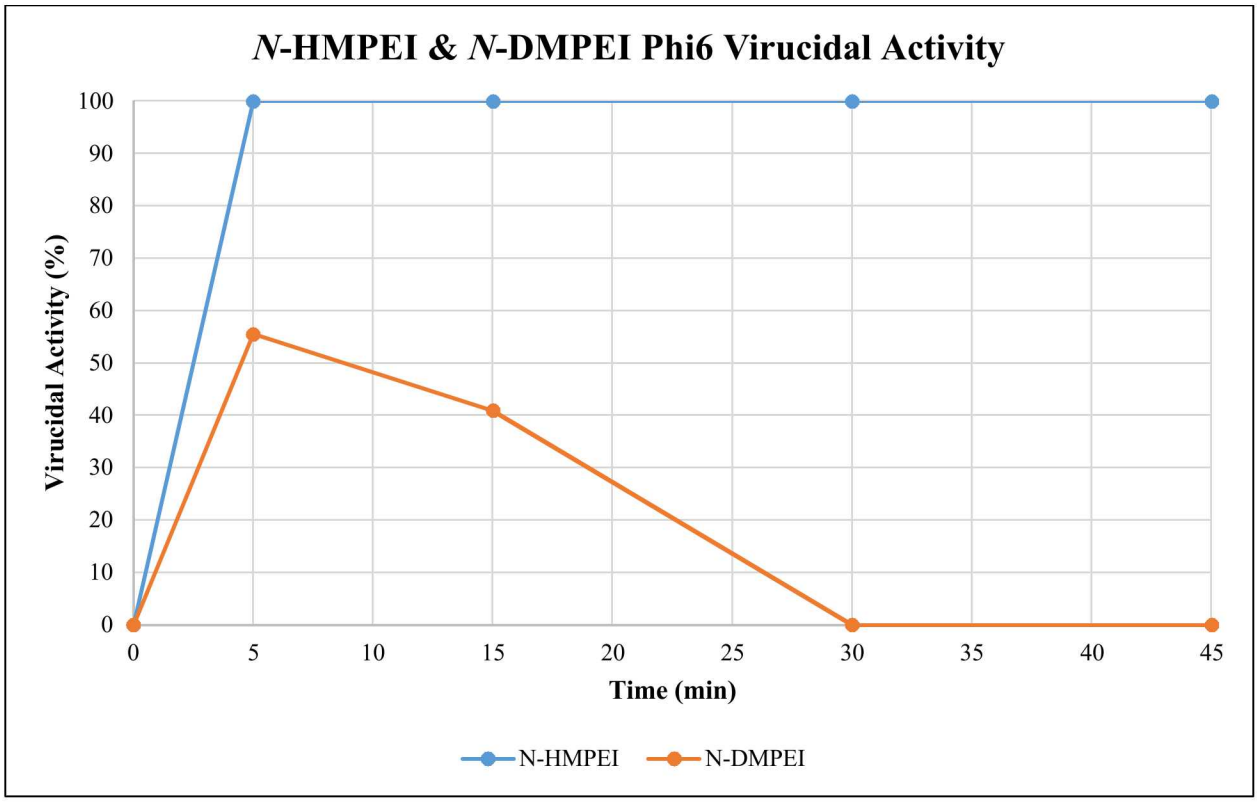


Figure 9. The time course of inactivation of phi 6 phage by a 24 well plate sprayed with branched 270 kDa *N*,*N*-alkyl-methyl PEI at room temperature.

Similar to the MS2 assays, treatment with *N*-HMPEI results showed complete inhibition of phi6 enveloped phage particles. Benzalkonium chloride also completely prevented the replication of phi6. Both the *N*-DMPEI sample and the no treatment control sample of just phi6 phage showed no impact to phage titer.

Conclusions

A sprayable polymer coating has been developed that shows essentially 100 % virucidal activity towards viruses with different structural properties. A 270 kDa branched polycationic PEI with hexyl and methyl alkyl groups off the polymer nitrogen atoms shows the greatest efficacy towards viral destruction. Spraying of aluminum surfaces with 1 wt % polymer solutions shows complete coverage of surface based on SEM/EDS, with some nanoscale cracking on some samples. Immersion of an N95 respirator into the polymer solutions shows good coating coverage on almost all layers of the filter material; however, the electrostatic neutralization between the polymer and electret material drastically reduces the filtration efficiency of the respirator. Finally, glass slides spray-coated with the polymer solution display tremendously high virucidal activity towards MS2 phage and phi6 phage in as little as 5 minutes after exposure. With these results, we expect these dilute organic polymer solutions to be effortlessly applied to solid substrates to yield surfaces that are immediately decontaminated and self-disinfecting for long periods of time.

Future Work

An assortment of material science is still needed to fully optimize the results obtained in this project. Lowering the concentration of the polymer and analyzing surface coverage and bio assays would deduce how much polymer is really needed in the solution to maintain anti-viral efficacy. Mixed solvent systems rather than individual ones could be useful in developing more-favorable coating morphological features based on evaporation rates. Furthermore, an unexpected experimental result was seen where the polymer dissolved in a mix of solvents, yet was insoluble in the individual solvents of the mixture; this observation could lead to valuable solubility information for other similar polymeric materials. Alternatively, the individual solvents determined from this work could all be sprayed and analyzed by SEM/EDS to get a suite of information for trend determinations. Finally, new synthetic pathways could be developed that generate new polymers with both anti-viral and physical elastomeric properties, yielding 1) greater durability and 2) more efficient surface coverage. Physical durability/longevity of all of these materials would also be valuable information to obtain as it would dictate how often reapplication of polymer would be needed.

For the N95 use-case, recharging of the coating using a coronal discharge could be attempted. Individual non-electret layers of the N95 could be coated during production (logistically difficult), or the spray method with the current polymer solution could be applied to the surface of the N95 and characterization performed.

The viral assays performed in this project ideally need to be continued with more repetitions to fully determine the average effects the coatings have on the viral surrogates. Furthermore, SARS-CoV-2 plaque assays would be the definitive test for use in eliminating COVID-19 contaminations.

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[2] Haldar, J.; An, D.; Álvarez de Cienfuegos, L.; Chen, J.; Klibanov, A.M. "Polymeric coatings that inactivate both influenza virus and pathogenic bacteria". *PNAS*, **2006,** *103*, 17667–17671.

APPENDIX A. SUPPLEMENTARY INFORMATION

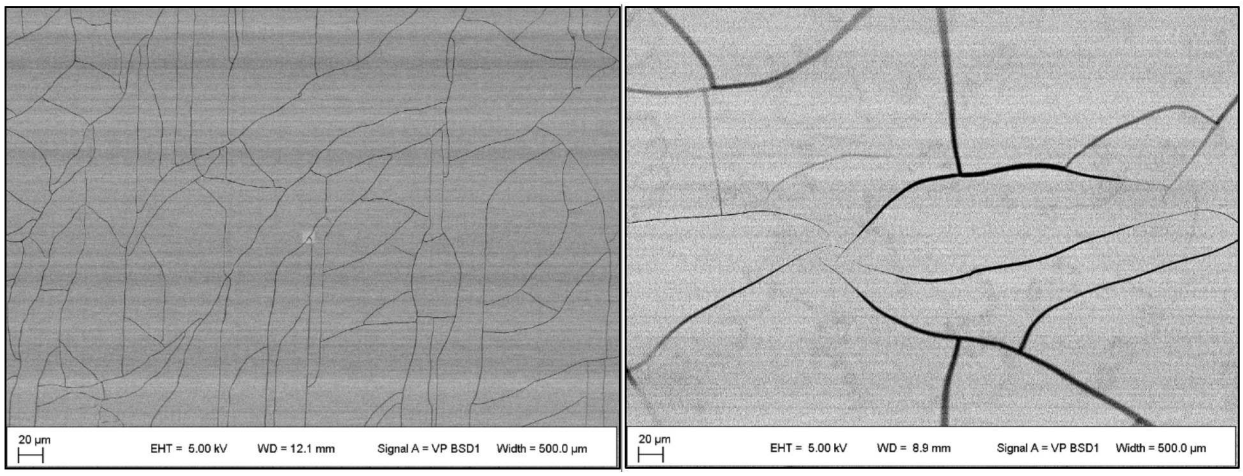

Figure S-1. SEM image of *N*-DMPEI (left) and *N*-HMPEI (right) applied from a 1 wt% solution in 2-butanol.

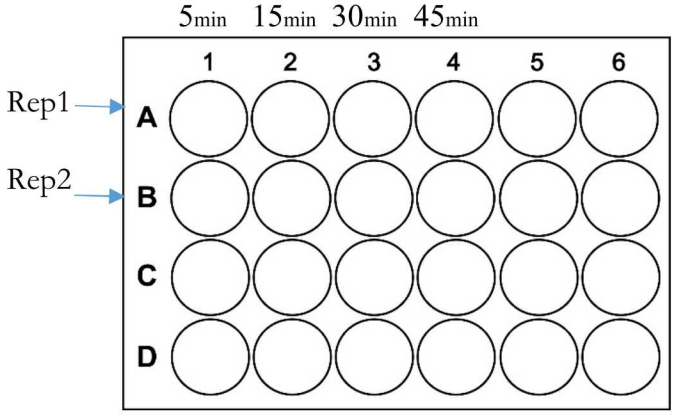

Figure S-2. Well plate setup for viral assays. Polymer solutions were sprayed inside wells to generate coating. Rep 1 = Repetition 1 and Rep 2 = Repetition 2.