Bleach-durable reactive silver ink coatings for anti-viral, repellent, and reusable textiles

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Abstract

There is a major need to improve reusable personal protective equipment (PPE) with anti-viral and liquid repelling functionality. Silver is a popular coating material to incorporate virus inactivation functionality to PPE. This work consists of utilizing reactive silver inks and low surface energy polymers to achieve anti-viral and and liquid repellency functionalities even after ultrasonic bleach washing. The virus inhibition and repellency functionalities of reactive silver inks and silver nanoparticles on fabric are measured before and after bleach washing. The potential of reusability for reactive silver inks is compared with traditional silver nanoparticle coatings on fabric.

Introduction

Personal protective equipment (PPE) such as gowns, masks, and scrubs are essential for protecting healthcare professionals from contact with bacteria and viruses that lead to infection. However, PPE may become contaminated during use and inadvertently contribute to the transmission of microbes. There is a need for textile coatings that provide PPE with better protection from infection by not only repelling fluids such as respiratory droplets, but killing or deactivating microbes. In particular, there is great interest in textile coatings that provide these functionalities in reusable PPE even after laundering.

Single-use or disposable PPE place a heavy burden on manufacturing supply chains that may lead to shortages during pandemics or times of great demand. Single use or disposable PPE also lead to large amounts of waste and environmental impact. Reusable PPE help alleviate burdens on manufacturing supply chains and decrease the amount of waste. Furthermore, they are more economical on a cost per use basis. However, one of the main challenges with reusable PPE is ensuring that they continue to provide protection from microbes after repeated use and decontamination. Reusable PPE are decontaminated after each use by laundering with bleach. Therefore, virus inactivating surface coatings for reusable PPE in medical settings must be able to withstand multiple laundering cycles in bleach solution.

Silver is a well-known anti-microbial agent. Previously, silver nanoparticles have been shown to inhibit the growth of yeast, Escherichia coli, and Staphylococcus aureus. More recently, silver nanoparticles have been demonstrated to have anti-viral properties against enveloped viruses such as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and human immunodeficiency viruses (HIV). Silver nanoparticles may interact with viral nucleic acids to disrupt serial viral infection and replication. Silver nanoparticles have been incorporated into a range of fabrics or masks due to their anti-microbial and anti-viral properties. However, nanoparticles on fabrics may have issues with being removed in the wash, resulting in the loss of any associated functionality.

may also pose a threat to aquatic organisms???? and bioaccumulate in the food chain.? Also, the efficacy of silver nanoparticles may be hindered after washing with oxidizing agents such as bleach or detergent.?

This work studies the virucidal activity of PET textiles coated with a silver amine complex ink? on herpes simplex viruses (HSV-1). PET fabric material is commonly used for medical and healthcare applications such as gowns, scrubs and caps.? HSV-1 is an enveloped virus about 160 nm in diameter that can cause cold sores and blinding herpetic eye infections.? The attachment and entry of HSV-1 into cells requires the interaction between the viral envelope glycoproteins and cell surface heparan sulfate (HS).? Silver mimics HS and competes for binding sites of the virus to the cell, inhibiting the virus from cell attachment and entry.??

The virus inactivating activity of reactive silver ink or 20 nm silver nanoparticles against HSV-1 in saline is investigated. Reactive silver ink in saline reduces HSV-1 activity by $1.6 \pm 0.9 \log_{10} \ (90.3 \pm 5.5\%)$ while nanoparticles in saline did not show significant activity reduction. The virus quantities of PET fabrics coated with silver ink or silver nanoparticles is compared. Once the silver ink is cured, the additional ingredients evaporate leaving a pure silver film. PET fabrics coated with 20 nm silver nanoparticles show a $1.1 \pm 0.6 \log_{10} \ (92.4 \pm 2.5\%, p < 0.05)$ log reduction in virus quantities compared to control fabric, while PET fabrics coated with reactive silver inks reduce virus quantities by $2.3 \pm 0.8 \log_{10} \ (99.5 \pm 3.0\%, p < 0.001)$. However, we observe the performance of bare silver fabric treatments can be significantly hindered after extended periods of bleach washing.

We show that the reactive silver ink provides for enhanced washing durability compared to nanoparticles. Reactive silver reduces *in-situ* on the textile and uniformly covers the microfibers of the fabric. This provides for better binding with the fabric compared to nanoparticles, which adhere to the fabric with weak van der Waals forces. An additional polydimethylsiloxane (PDMS) thin film can protect the silver layer and increase the liquid repellent properties or the fabric to improve the durability of the functionality after de-

contamination by bleach washing. PET fabric samples coated with reactive silver ink and PDMS reduce virus quantities by $1.7 \pm 0.2 \log_{10} (98.2 \pm 0.8\%, p < 0.01)$ after 300 minutes of ultrasonic bleach washing. We demonstrate the combination of reactive silver inks with PDMS as a PET fabric treatment that adds durable superhydrophobic and virus reducing properties for reusable PPE applications.

Results and discussion

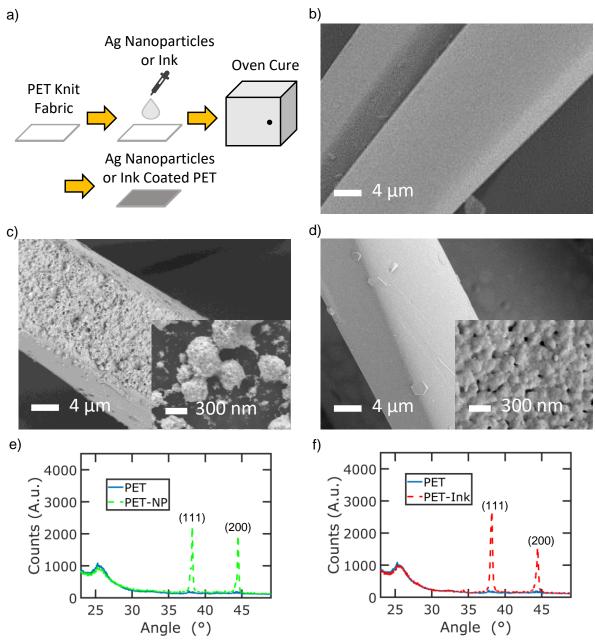


Figure 1: a) Schematic of silver treatment on PET fabric (b-d) SEM images of b) untreated PET c) PET-NP and d) PET-Ink. e-f) XRD spectra of e) PET-NP and f) PET-Ink samples.

Fig. ?? shows the results of coating the PET fabric with silver nanoparticles or ink. The PET fabrics are coated by drop casting and then thermally cured (Fig. ??a). Square fabric samples of 0.5 in. by 0.5 in are coated with either silver nanoparticles or ink by drop casting, followed by curing in an oven for 1 hour at 120 °C. An equal amount of silver by weight (0.2

mg) was added to each fabric. The untreated PET knit fabric consist of microfibers about 12-15 µm in diameter (Fig. ??b). The 20 nm silver nanoparticles (PET-NP) aggregate and attach non-uniformly over the surface of the PET (Fig. ??c). In contrast, PET fibers coated with silver ink (PET-Ink) is observed to be more uniform around the PET fibers (Fig. ??d). X-ray diffraction (XRD) is performed on the coated samples (Fig. ??e-f). Distinct peaks are observed at $2\theta = 38.2^{\circ}$ and 44.5° corresponding to the (111) and (200) crystal planes. The XRD analysis confirms the presence of poly-crystalline silver on PET for nanoparticle (Fig. ??e) and ink (Fig. ??f) coatings, respectively.

a)

Sample	Water Static Contact Angle	Advancing Contact Angle	Receding Contact Angle	Water Hysteresis	Water Breakthrough Pressure
PET	0°	~	~	~	~
PET-NP	129 ± 2°	138 ± 5°	121 ± 5°	18 ± 4°	250 ± 20 Pa
PET-Ink	139 ± 5°	146 ± 6°	130 ± 8°	16 ± 2°	332 ± 17 Pa

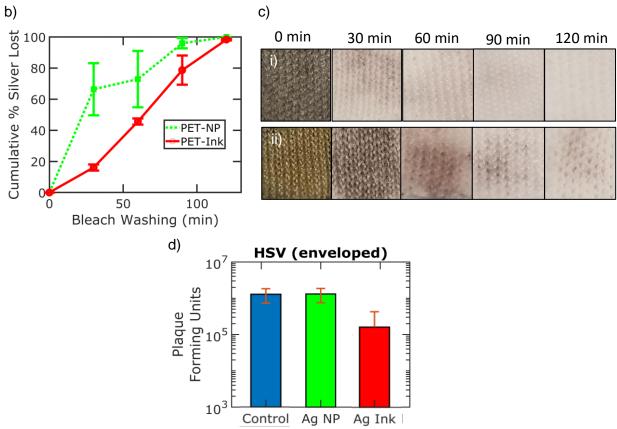


Figure 2: a) Wetting characterization for PET, PET-NP, and PET-Ink samples. b) Silver leaching analysis as a function of bleach washing time c) Optical images of (i) PET-NP and (ii) PET-Ink samples as a function of bleach washing time. d) Virus inhibition assay results of silver treatments in solution.

Fig. ??a shows the wetting properties of coated PET samples and virus inhibition properties of the silver coatings. Untreated PET is fully wetted by water. The water static contact angle and hysteresis of PET-NP is $129 \pm 2^{\circ}$ and $18 \pm 4^{\circ}$, respectively. The breakthrough pressure is estimated to be 250 ± 20 Pa based on the water droplet radius when the droplet transitions from a Cassie-Baxter state to a Wenzel state. The water static contact angle

and hysteresis of PET-Ink is $139 \pm 5^{\circ}$ and $16 \pm 2^{\circ}$, respectively. The breakthrough pressure is estimated to be 332 ± 17 Pa. Both coated PET samples show similar hydrophobic wetting properties due to the nanoscale roughness on microfibers.

Bleach washing experiments were performed to assess the durability of silver. The cumulative percentage of silver that came off coated samples during bleach washing is shown in Fig. ??b. The silver concentration from the affluent of each 30 minute washing cycle was measured using inductively coupled plasma mass spectrometry (ICP-MS). PET-NP samples lose $62 \pm 9\%$ of the coated silver after the first 30 minutes of washing. PET-Ink samples lose silver at a slower rate due to the more uniform coating and stronger attachment to the PET from the silver diamine complex. Both treatments lose at least 99% of the silver by 120 minutes of washing. Fig. ??c shows representative images of the (i) PET-NP and (ii) PET-Ink samples as a function of bleach washing cycles. Bleach oxidizes silver, which degrades the intrinsic properties. It is evident that silver treated fabrics need additional surface treatment for reuse in bleach washing processes.

Virus assays in saline are conducted by mixing silver ink or silver nanoparticles (0.1%) in phosphate buffered saline (PBS) with HSV-1 in PBS for 1 hour, followed by centrifuging, removing the supernatant, and quantifying the amount of virus remaining. Fig. ??d shows the results of inactivation experiments of nanoparticle or reactive ink-coated PET on enveloped HSV-1 in phosphate buffered saline (PBS) at a concentration of 1 mg per mL (0.1%). While the 20 nm silver nanoparticles do not show HSV-1 inactivation with a $0.0 \pm 0.1 \log_{10}$ difference, the silver ink shows HSV-1 inactivation in PBS by $1.6 \pm 0.9 \log_{10}$ (90.3 \pm 5.5%) difference. Previous reports have suggested that virus inactivation is stronger with smaller diameter silver nanoparticles (\leq 15 nm) and it is possible the nanoparticles are too large to cover the binding sites of HSV-1 to inhibit cell entry. Also, the silver ink has additional components such as octylamine (5-10%) and ammonium formate (1-5%) which may contribute to the virus reduction.

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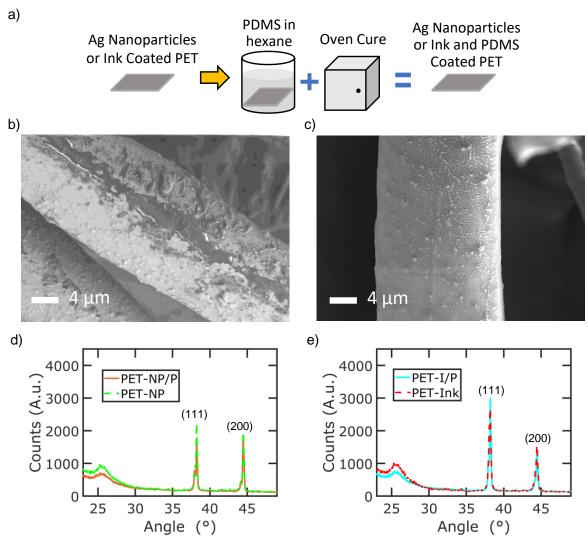


Figure 3: a) Schematic of PDMS treatment for silver coated PET samples (b-c) SEM images of treated fiber of b) PET-NP/P and c) PET-I/P samples. d-e) XRD spectra of fabric samples before and after PDMS coating for d) PET-NP/P with PET-NP and e) PET-I/P with PET-Ink samples.

A PDMS post treatment is added to the silver coated samples to improve the wash durability. PDMS is a low surface energy polymer that offers liquid repellent properties to fabrics. Also, PDMS does not react heavily with bleach to add chemical resistance. Both PET-NP and PET-Ink samples are dipped in PDMS (1:10, Sylgard) and cured in an oven at 150 °C for two hours to make PET-NP/P and PET-I/P samples. Fig. ?? shows the characterization of PET-NP/P and PET-I/P samples. A schematic of the PDMS treatment process is depicted in Fig. ??a. SEM imaging shows the physical morphology of microfibers for PET-NP/P (Fig. ??b) and PET-I/P (Fig. ??c) samples. The PDMS layer can be observed

on the microfibers for both treatments. XRD spectroscopy identifies the crystalline phases present on PET-NP/P (Fig. ??d) and PET-I/P (Fig. ??e) microfibers before and after PDMS coating. The distinct silver peaks of (111) and (200) are still present for both samples after the PDMS coating, confirming the presence of silver on the samples. The PDMS coating adds broader peaks at $2\theta = 22-27$ °due to the amorphous polymer structure.

a)										
	Sample	Water Static Contact Angle	Advancing Contact Angle	Receding Contact Angle	Water Hysteresis	Water Breakthrough Pressure				
	PET-NP/P	145 ± 4°	145 ± 2°	133 ± 3°	13 ± 4°	388 ± 21 Pa				
	PET-I/P	156 ± 1°	156 ± 1°	148 ± 2°	7 ± 3°	490 ± 65 Pa				
b)		c)							

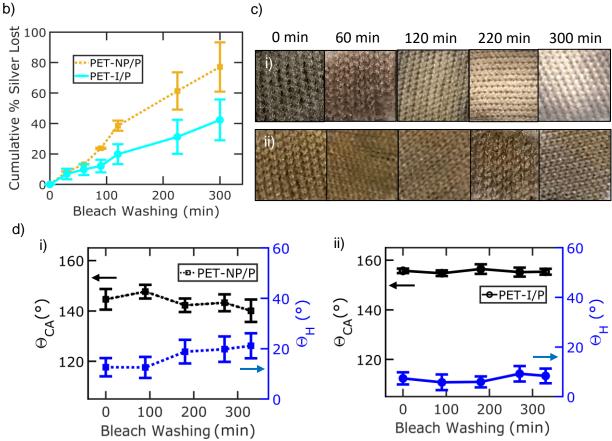


Figure 4: a) Wetting characterization for PET-NP/P and PET-I/P samples b) Silver leaching analysis as a function of bleach washing time c) Optical images of (i) PET-NP/P and (ii) PET-I/P samples as a function of bleach washing time d) Water contact angle (black) and hysteresis (blue) as a function of ultrasonic bleach washing time for (i) PET-NP/P and (ii) PET-I/P

Fig. ??a shows the wetting properties of PET-NP/P and PET-I/P samples. The water static contact angle and hysteresis of PET-NP/P is $145 \pm 4^{\circ}$ and $13 \pm 4^{\circ}$, respectively. The breakthrough pressure is 388 ± 21 Pa from an estimated droplet radius of 370 µm at breakthrough. The water static contact angle and hysteresis of PET-I/P is $156 \pm 1^{\circ}$ and $7 \pm 3^{\circ}$, respectively. The breakthrough pressure is 490 ± 65 Pa from an estimated droplet radius of 294 µm at breakthrough. The PDMS layer significantly improves the liquid repellency properties of the fabrics for both treatments. PET-I/P demonstrates more water repellency than PET-NP/P due to the uniform, nanoscale roughness of the ink along all microfibers. PET-NP/P samples are not uniformly covered by silver; therefore, there exists more fabric surface area without nanoroughness which leads to more liquid penetration.

The cumulative percentage of silver that came off PDMS coated samples during bleach washing is shown in Fig. ??b. The additional PDMS layer of PET-NP/P and PET-I/P samples significantly improves the retention of silver compared to PET-NP and PET-Ink samples. The PDMS layer protects the silver layer from bleach oxidation and extends the washing durability of the samples. PET-NP/P samples lose more silver compared to PET-I/P samples. PET-I/P samples lose silver at a slower rate due to the stronger liquid repellency properties and higher breakthrough pressure. PET-I/P maintains about $42 \pm 17\%$ more silver than PET-NP/P after 300 ultrasonic bleach washing minutes. Fig. ??c shows representative images from (i) PET-NP/P and (ii) PET-I/P of the silver loss as a function of bleach washing cycles. PET-I/P retain color much better than PET-NP/P samples, which become stained from the bleach oxidizing and removing the silver.

Fig. ??d illustrates the water static contact angle and hysteresis of (i) PET-NP/P and (ii) PET-I/P samples as a function of bleach washing time. The average change in static water contact angle and hysteresis for PET-NP/P is $7 \pm 2^{\circ}$ and $9 \pm 1^{\circ}$ after 300 minutes of bleach washing. The average change in static water contact angle and hysteresis for PET-I/P is $1 \pm 1^{\circ}$ and $2 \pm 1^{\circ}$ after 300 minutes of bleach washing. The PDMS layer helps retain silver on PET samples during bleach washing; however, the PET-I/P coated samples show better

performance and durability due to the stable liquid repelling properties.

In this work, we measure the quantity of herpes simplex virus (HSV-1, enveloped) on fabrics, using standard plaque forming unit (PFU) assays, for samples that are unwashed, bleach washed for 100 minutes and bleach washed for 300 minutes samples. Virus assays on fabric are conducted by submerging fabrics in virus/PBS while rocking for 1 hour, then samples are removed and placed in fresh PBS. Lastly, samples are sonicated at low power to remove viruses that adhered on the samples. The PBS liquid afterwards is used to quantify the amount of virus present on the samples. Mann-Whitney are utilized to offer a degree of statistical certainty that the fabric treatment reduces the amount of virus particles on the fabric compared to controls. Asterisks corresponds to the level of certainty that the treated samples are the same as the controls where one asterisk corresponds to $p \leq 0.05$, two for p < 0.01 and three for p < 0.001.

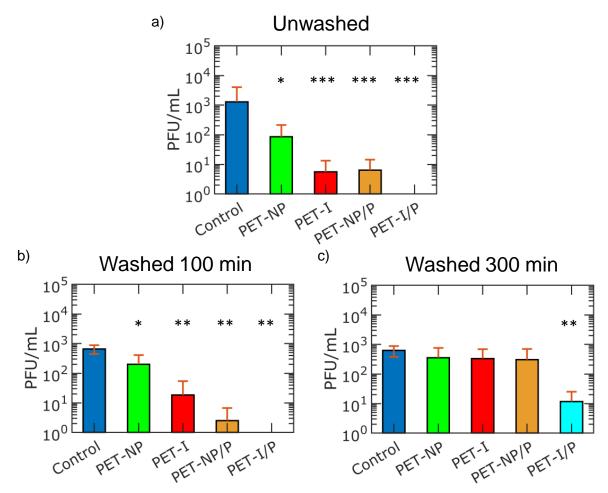


Figure 5: a) HSV-1 viral quantities for unwashed fabric samples b) HSV-1 viral quantities for fabric samples after 100 minutes of bleach ultrasonic washing c) HSV-1 viral quantities for fabric samples after 300 minutes of bleach ultrasonic washing.

The amount of virus on fabric samples before and after ultrasonic bleach washing is shown in Fig. ??. Fig. ??a shows the amount of active virus on unwashed fabric samples. PET-NP, PET-Ink, PET-NP/P and PET-I/P samples show a reduction of HSV-1 by $1.1 \pm 0.6 \log_{10} (92.4 \pm 2.5\%, p < 0.05)$, $2.3 \pm 0.8 \log_{10} (99.5 \pm 3.0\%, p < 0.001)$, $2.3 \pm 0.8 \log_{10} (99.5 \pm 3.1\%, p < 0.001)$ and $2.8 \pm 0.2 \log_{10} (99.8 \pm 0.9\%, p < 0.001)$ PFU per mL, respectively. PDMS swells in the presence of liquid which potentially allows silver to slowly diffuse. ? ? The reduction of activated virus is from a combination of liquid repellency and virus inactivation.

The amount of virus on fabric samples after 100 minutes of bleach washing is shown in Fig. ??b. PET-NP, PET-I, PET-NP/P and PET-I/P samples show a reduction of HSV-1 by

 $0.6 \pm 0.4 \log_{10} (74.8 \pm 2.0\%, p < 0.05), 1.6 \pm 0.4 \log_{10} (97.2 \pm 1.9\%, p < 0.01), 2.4 \pm 0.3 \log_{10} (99.6 \pm 1.0\%, p < 0.01)$ and $2.8 \pm 0.1 \log_{10} (99.8 \pm 0.4\%, p < 0.01)$ PFU per mL, respectively. Samples without the PDMS layer lose some functionality as significant amounts of the silver is removed from washing. Samples with the PDMS layer maintain liquid repellency and silver after 100 minutes of bleach washing due to the chemical resistance of PDMS which retains the silver layer.

Lastly, the amount of virus on fabric samples after 300 minutes of bleach washing is shown in Fig. ??c. PET-NP, PET-I samples and PET-NP/P samples no longer show statistically significant reductions in HSV-1 after this extended bleach washing time from a lack of silver and liquid repellency properties. PET-I/P samples after 300 minutes of bleach washing still show a reduction of virus by $1.7\pm0.2\log_{10}$ (98.2±0.8%, p < 0.01) PFU per mL by the liquid repellency and virus inactivation properties. Overall, the PET-I/P treated fabrics reduce the amount of active virus, even after 300 minutes of bleach washing. The ink coating covers the fibers more uniformly and the PDMS coating adds silver retention and liquid repellent properties for reusable, anti-viral performance.

Conclusion

There is a need for reusable, functional PPE to improve the public health safety against viral infections. This work demonstrates how silver ink and low surface energy polymers can be used to repel liquid and inactivate viruses on polyester fabric with long lasting functionality. The fabric treatment fully repels water before and after bleach washing. Most importantly, the fabric treatment shows reductions in enveloped virus quantities by about 2 logs compared to controls before and after bleach washing. This work demonstrates a durable, superhydrophobic, virus inactivating functionality on common fabric for reusable PPE applications.

Experimental

Sample Preparation

Materials: PET knit wipes were purchased from Anticon. Acetone (99.5%), methanol (99.9%) and isopropyl alcohol (99.5%) were bought from VWR. PBS, FBS, PDMS (Sylgard) and were bought from Sigma-Aldrich. Reactive silver inks (720 series) was provided by Electroninks. Silver nanoparticles (20 nm, 1mg/mL Citrate) were purchased from NanoComposix. Household bleach was purchased from Giant Eagle. Deionized water was used from a Millipore Academic A10 system with total organic carbon below 40 ppb. Herpes simplex virus were obtained from the American Type Culture Collection (ATCC), Manassas, VA. Virus stocks in PBS were prepared with A549 human lung carcinoma cells and were cesium chloride centrifuge purified to remove any cellular and FBS proteins. The virus stocks were diluted in sterile PBS to the experimental titers used.

Sample Fabrication: 0.5 in. by 0.5 in. square samples were cut from PET fabric. All samples were rinsed with acetone, methanol and isopropyl alcohol and dried with nitrogen to eliminate possible contaminants. An equal amount of 0.2mg of silver was added to fabric samples, confirmed by microscale. Silver ink was added by drop casting onto untreated PET fabrics followed by oven curing for 1 hour at 120 °C to make PET-Ink samples. Silver nanoparticles was added by drop casting onto untreated PET fabrics followed by oven curing for 1 hour at 120 °C to make PET-NP samples. Additional PET-Ink and PET-NP samples were treated with a PDMS layer by soaking samples in PDMS (Sylgard) at 1:10 curing agent ratio and curing for two hours at 150 °C to make PET-I/P and PET-NP/P samples, respectively.

Sample Characterization

The physical morphologies of samples were characterized by scanning electron microscopy (SEM, Zeiss Sigma 500 VP) at 5kV. For SEM imaging, all samples were sputter coated with

10 nm gold/palladium (80:20) using a sputter coater (Denton). The presence of silver was confirmed used X-ray diffraction (XRD).

Static, advancing and receding contact angle measurements were taken in ambient air at 22–25°C and 20–30% relative humidity using an optical tensiometer (Attension, 811 Theta). 5 µL droplets at 25°C for all test liquids were used for all wetting measurements. The hysteresis was tabulated for each treatment after measuring the advancing and receding contact angles during syringe-controlled water dispersion and withdrawal, respectively.

Breakthrough pressure was measured by observing the contact angle and volume while a water droplet evaporates. When the droplet transitioned from Cassie-Baxter to Wenzel state the diameter of the droplet was tabulated to calculate the breakthrough pressure.

Durability Testing

Washing cycles were performed using a Powersonic P230 Ultrasonic Cleaner (Crest) under ASTM G131-96 standards for washing materials by ultrasonic techniques. 10% bleach washing solution was prepared in a 10 mL test tube to create a highly efficient washing solution. Samples were submerged in bleach solution in Eppendorf tubes and ultrasonicated for 30 minutes at 80 W and 54°C to complete one wash cycle. Afterwards, samples were dried in ambient temperature before testing.

Silver leaching analysis was conducted using inductively coupled plasma-mass spectrophotometer (ICP-MS). Wash affluent was collected as samples were subject to bleach washing cycles. Then, samples were diluted for ICP-MS by mixing 9.8 mL of water (2% nitrate solution), 10 μ L of standard and 20 μ L of wash affluent and taking the weights using a microscale. The weights were used to calculate dilution factors for ICP-MS results.

Virus Assays

Virus inactivation assays were performed by mixing silver nanoparticles or silver ink with herpes virus in PBS at final concentration of 1 mg per mL (0.1%). Solutions were vortexed

and incubated for 1 hour at room temperature. After 1 hour of incubation, 500 µl of ice-cold tissue culture media containing 20% FBS was added, then vortexed and centrifuged for 1 minute in the Eppendorf centrifuge to pellet any silver. The supernatants were removed and plated with 0.1 ml of serial 10-fold dilutions in duplicate onto A549 monolayers in 24 well multiplates. The virus was absorbed for 1-3 hours after which the wells were filled with 1 ml of tissue culture media containing 0.5% methyl cellulose. Plates were incubated for 5-6 days in 5% CO2 and then fixed and stained with 0.5% gentian violet solution containing formalin and the plaques were quantified.

Assays on Fabrics: Fabric samples were completely submerged in 0.4 mL of virus/PBS in Eppendorf tubes with moderate shaking for 60 minutes (Stoval Belly Dancer, level 5) at room temperature. After shaking, samples were gingerly rinsed once in sterile PBS and submerged in Eppendorf tubes with 0.4 mL of PBS. Then, virions attached on the surface were removed from the samples into PBS by sonication at power 3 for 10 seconds (Qsonica, Model Q55) within the tubes. The remaining PBS was used to quantify plaques.

Virus titers (PFU per mL) for herpes simplex virus were determined using standard plaque assay with A549 human lung carcinoma cells prepared in 24-well tissue culture plates. After 6-7 days incubation at 37°C in 5% CO2, the cells were fixed and stained with gentian violet prepared in formalin, the number of plaques per well were counted under a dissecting microscope, and viral plaque forming unit titers were calculated. Finally, Mann-Whitney U-tests rejected the null hypothesis at p values < 0.05 for all comparisons between control and treated samples.

Acknowledgement

The authors thank Dr. Ke Ren for his statistical expertise.

Supporting Information Available

Virus inactivation experiment results, goniometer images of static water contact angles of fabric samples, XRD spectra and SEM images of PDMS treated fabric samples after bleach washing.

References

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