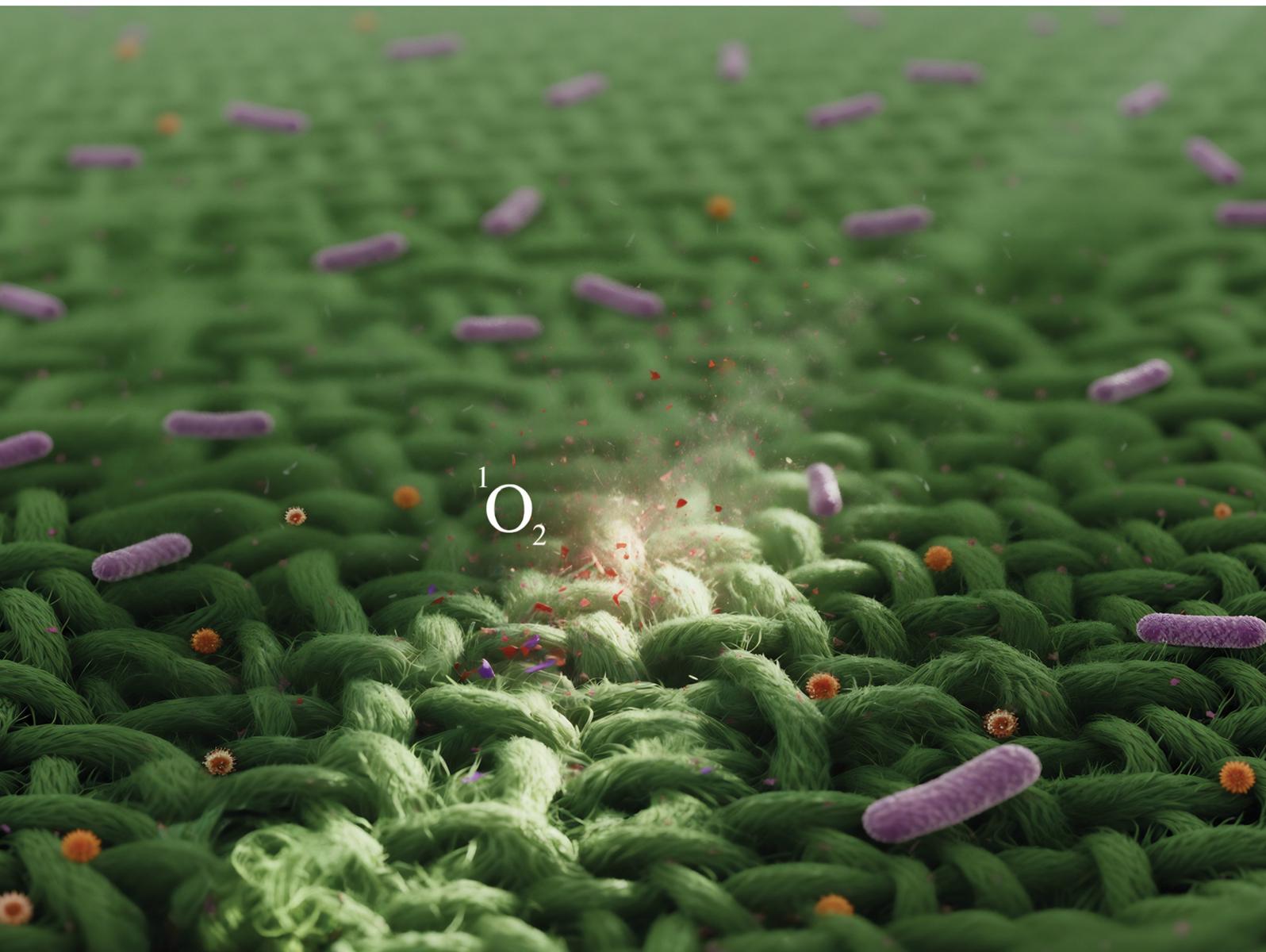


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PAPER

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Photo-activated antimicrobial textiles and paper using non-covalently adsorbed zinc phthalocyanines

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Effective personal protective equipment (PPE) and operational clothing and equipment (OCE) are essential to protect healthcare workers and the population at large from infectious microbes, as highlighted by the COVID-19 pandemic. The demand for effective and technologically advanced PPE/OCE is high due to the threat of new viral pandemics and growing antibiotic resistance. PPE and OCE typically act as physical barriers to respiratory droplets and other body fluids and thus are limited in their ability to protect users. Here we sought to develop PPE/OCE materials that actively kill bacteria and inactivate viruses upon contact. Photoactivatable metallophthalocyanines (PcMs) have antimicrobial properties and are more cost-effective and less toxic than metals and nanoparticles as surface treatments. We identified several PcMs with strong in-solution activity for both Gram-positive and Gram-negative bacterial pathogens and diverse viral pathogens. These PcMs were used to dye textiles and paper under mild conditions. Upon photoactivation cotton and paper dyed with the water-soluble PcM **RLP068/OTf** produce a 5–6- \log_{10} reduction in viable Gram-positive bacteria, a 3- \log_{10} reduction in Gram-negative bacteria, and a 1–2- \log_{10} reduction in active enveloped viruses. A combination of **RLP068/OTf** and the water-insoluble PcM **4Bulmid/OTf** produced a 4–5- \log_{10} reduction in both Gram-positive and Gram-negative bacteria. Leaching of these compounds into the surrounding medium is minimal, suggesting their microbicidal activity results from direct contact. The self-sterilizing textiles identified here are promising raw materials for low-cost PPE and OPE in preparation for pandemics and for everyday protection in sectors at high risk from pathogen transmission.

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Introduction

The World Health Organization recognizes viral pandemics and antimicrobial resistance as major threats to public health worldwide.¹ Transmission of microbial pathogens is particularly problematic in locations with potentially high rates of infection, such as hospitals and care homes, and where people live in close quarters, such as prisons, army barracks, and naval

and cruise ships. The 2020 worldwide COVID-19 pandemic highlighted the need for pandemic preparedness, including robust methods to minimize microbial transmission in patient care and public settings, and for improved personal protective equipment (PPE) for healthcare personnel, frontline workers and the general public. Reusable PPE would reduce the overall demand, and PPE with antimicrobial properties can serve not only as protective physical barriers for aerosolized pathogens but can also kill or inactivate infectious microbes, thereby limiting their transmission.

Common methods used to clean PPE for reuse in healthcare settings include exposure to disinfectants such as ethanol, quaternary ammonium compounds (e.g., Lysol™), bleach and hydrogen peroxide.² Such treatments are time-consuming, may generate significant waste, and can degrade materials over time or leave residues.³ Other sterilization methods such as heat/steam and ultraviolet (UV) light, and gamma radiation, can

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break down components of PPE such as plastic, and UV exposure of all PPE surfaces is not always possible.⁴ Moreover, such exposure typically requires a controlled setting, not at the point of use. Self-sterilizing PPE containing antimicrobial agents reduce or eliminate the need for decontamination and provide added protection to the user. Antibacterial and antiviral agents like copper and silver have been used in coatings and incorporated into PPE materials to improve their protective properties and extend their use, but the use of these metals is limited because of their cost. Nanoparticles made from copper, silver, or other nanomaterials are effective anti-microbial agents⁵ but are not desirable due to their toxicity and potential diffusion into the environment.⁶

Macrocyclic photoactive compounds like porphyrins and phthalocyanines have shown much promise as broad-spectrum antimicrobial agents for PPE. These compounds can be photosensitized by visible light, producing singlet oxygen that oxidizes macromolecules such as proteins and lipids on the microbial surface.^{7–11} Since these molecules non-specifically target critical structural components of bacteria and viruses, such as membranes, they are less likely to drive resistance. They have proven to be safe¹² and are used internally in photodynamic therapies.^{13,14} Phthalocyanine derivatives are effective against both Gram-positive and Gram-negative bacteria and viruses in solution.^{7–10,15–17} These compounds have been immobilized on materials (*e.g.* paper, cotton, synthetic polymers) *via* covalent and non-covalent means to produce self-sterilizing materials for PPEs.^{12,18,19} Covalent attachment prevents diffusion of the compounds into the surrounding medium (leaching) but can be costly at large scale. Non-covalent attachment by dyeing is simple and cost-effective but can result in leaching. Cuthbert *et al.* covalently cross-linked zinc porphyrin to the surface of a polypropylene textile used in filters of N95 respirators. This treated material, when incubated with influenza A virus in solution and exposed to high intensity visible light, reduced viral activity by 99.99% (4-log₁₀ reduction).²⁰ Non-covalent attachment of zinc tetrakis(*N*-methylpyridinium-4-yl) tetraiodide phthalocyanine (LASU) to filter paper by dyeing resulted in a ~4-log reduction in colony-forming units (CFUs) for the Gram-negative

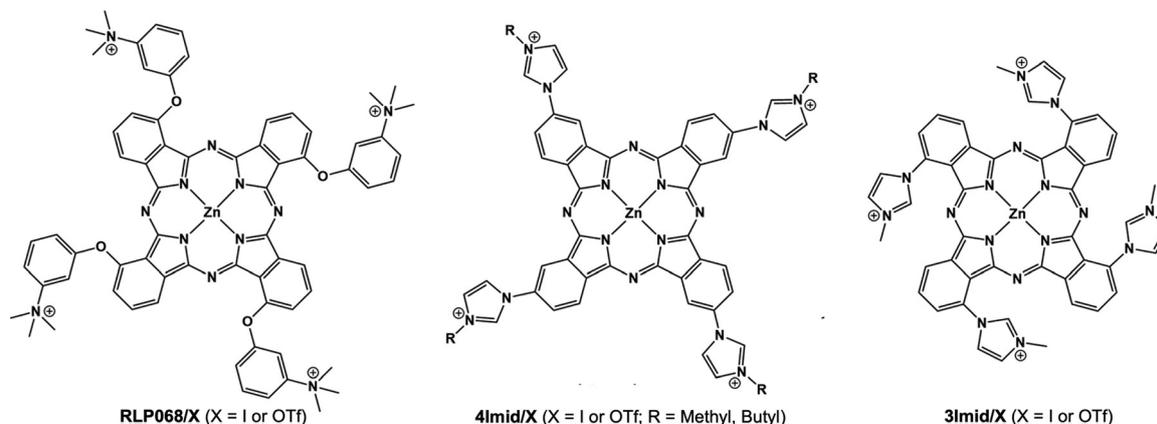
bacteria *Escherichia coli* and *Acinetobacter baylyi* with minimal leaching.²¹ Cotton dyed with LASU showed a 3–4-log reduction in coronavirus HCoV-229E titer upon activation with common light sources.¹² The related zinc tetra(4-*N*-methylpyridyl)porphyrin was incorporated into a thermoplastic elastomer by solution-casting and melt-pressing at 140 °C; this material reduced CFUs by 3–6 logs for several bacterial pathogens, and inactivated viral pathogens by 3–5 logs.⁶

Here we report the synthesis of thermostable derivatives of the metallophthalocyanine (PcM) **RLP068** known to have antimicrobial activity,¹⁵ and of imidazole-substituted PcM systems, the *N*-ethyl versions of which were reported to be antimicrobial.^{22,23} These compounds were used to dye textiles and paper under mild, cost-effective and scalable conditions. The dyed materials were tested against a panel of human pathogens and shown to kill both Gram-positive and Gram-negative bacteria and to inactivate enveloped viruses upon direct contact with no detectable leaching. These inexpensively produced self-sterilizing materials can be used in disposable and broadly protective PPE, extending their lifespan and reducing transmission of microbial pathogens.

Results

Synthesis and thermostability of photoactivatable metallophthalocyanines **RLP068/I**, **RLP068/OTf**, **3Imid/I**, **4Imid/I**

A series of tetra- (Scheme 1) and octa-cationic zinc PcMs were prepared. Some of these compounds were reported previously and have recognized anti-microbial activity *via* photo-generated singlet-oxygen,^{15,22,23} while others are new analogues. For example, (Me₂NPhO)₄PcZn and **RLP068/I** were synthesized as described¹⁵ where (Me₂NPhO)₄PcZn was cyclized and metalated in one step from the respective phthalonitrile and zinc acetate, and **RLP068/I** was prepared *via* the reaction of iodomethane and (Me₂NPhO)₄PcZn. Imidazole-substituted zinc phthalocyanines, where four imidazole substituents are located on the non-peripheral



Scheme 1 Chemical structures of PcMs utilized herein. Anions (I⁻ or OTf⁻) are not shown. One of several possible regioisomers is shown in each case. (**3Imid** may be a single isomer due to steric factors).



α -positions (**3Imid**) or the peripheral β -positions (**4Imid**) or the recently reported octa-substituted version in the β -position (**8-4Imid/I**), were prepared from direct cyclization of the appropriate phthalonitrile; methylation with iodomethane provided the tetra- or octa-methylated versions accordingly (**3Imid/I**, **4Imid/I** or **8-4Imid/I** respectively).^{22,24} To improve their thermal stability the iodide anions in **RLP068/I**, **4Imid/I** and **4DMAE/I** were substituted with triflates *via* salt metathesis with silver triflate in methanol, to form **RLP068/OTf**, **4Imid/OTf**, and **4DMAE/OTf**; other anions including nitrate and SbF_6^- were also surveyed to assess their impact on thermal stability of the **RLP068/I** system. The silver iodide byproduct in these reactions was readily suspended and removed *via* repeated centrifugation and decantation. **RLP068/OTf** can also be synthesized directly from $(\text{Me}_2\text{NPhO})_4\text{PcZn}$ and methyl triflate, eliminating a synthetic step, the use of a silver salt, and the purification steps to remove the silver iodide. The ionic materials (typically in the form of iodide or triflate salts) are soluble in water and could be applied as dyes in aqueous solutions. The water-insoluble **4Bulmid/OTf** was prepared by reaction of *n*-butyl iodide with **4Imid**²⁵ followed by salt metathesis with AgOTf . Synthetic and characterization details are provided in the SI.

Thermo-gravimetric analysis (TGA) was performed to identify compounds capable of withstanding potentially high dyeing and washing temperatures (at least 120 °C). The previously reported PcM **RLP068/I** sheds MeI at 80 °C, rendering it unsuitable as a dye (Fig. 1). As noted above, to identify a more thermostable **RLP068/X** PcM and overcome the impact of the low-boiling point of MeI , several salts with different anions (X) were prepared *via* AgX metathesis with **RLP068/I** (generating insoluble AgI and **RLP068/X**; X = OTf^- , SbF_6^- , NO_3^-). Although the SbF_6^- salt is unsuitable for biological applications due to its inherent toxicity it was included to explore the anion's impact on the thermal stability of **RLP068/X**. Of these, the triflate salt **RLP068/OTf** showed the highest thermal stability by TGA, with an onset decomposition temperature of 340 °C, which is

comparable to the precursor $(\text{Me}_2\text{NPhO})_4\text{PcZn}$ (~ 400 °C) (Fig. 1 and Fig. S1). The high thermostability of **RLP068/OTf**, together with its water solubility, makes it an attractive dye for antimicrobial studies. Moreover, the remarkable increase in thermostability by simple anion substitution presents a novel method to drastically tune the thermal properties of cationic PcMs.

RLP068, 3Imid, 4Imid phthalocyanines are active toward *S. aureus* and *P. aeruginosa* in solution

To identify promising candidates for use in antimicrobial textiles we first tested our panel of water-soluble PcM solutions for their ability to kill Gram-negative and Gram-positive bacteria and to inactivate viruses. For the antibacterial assays the Gram-positive species *S. aureus* and the Gram-negative species *P. aeruginosa* and *E. coli* were tested. Bacteria were mixed with varying concentrations of PcMs and exposed for 1 h to common indoor light sources: halogen, warm LED or fluorescent light. Cells were then serially diluted, plated on nutrient agar and grown overnight to determine the number of surviving cells. For our purposes we define a minimal inhibitory concentration (MIC) as the PcM concentration producing a 6-log reduction in CFUs, which represents the limit of detection (LOD) for our assay. Of the compounds tested, three PcMs, **RLP068/X**, **3Imid/I** and **4Imid/X**, showed substantial antibacterial activity, both as X = OTf and I salts (Table 1 and Fig. S2). These compounds showed greater efficacy for the Gram-positive bacterium *S. aureus*, with MICs of 1 μM , than for the Gram-negative species *P. aeruginosa* and *E. coli*, which required ~ 10 -fold higher concentrations to completely inhibit bacterial growth. **RLP068/I** and **RLP068/OTf** showed the greatest efficacy, with several logs of reduction even at 0.1 μM for *S. aureus* with halogen light activation (Fig. S2). Warm LED and fluorescent lights are approximately as effective as halogen lights for activating **RLP068/X** toward *S. aureus* but are less effective at activating the imidazole PcMs **3Imid/I** and **4Imid/X**, requiring ~ 10 -fold higher concentrations to inhibit growth. At high

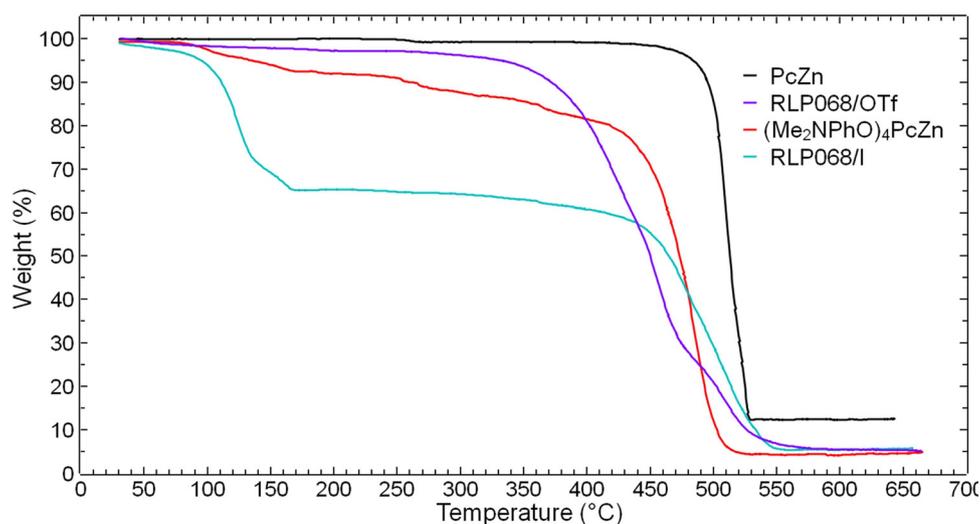


Fig. 1 Thermogravimetric analysis of **RLP068**-based PcZn complexes (neutral system and **RLP068/X**, where X = I, OTf) and unsubstituted PcZn as a reference.



Table 1 Antimicrobial activity of phthalocyanines in solution

Compound	Antibacterial activity			Antiviral activity					
	Gram-positive		Gram-negative	Enveloped			Non-enveloped		
	<i>S. aureus</i>		<i>P. aerug</i>	<i>E. coli</i>	HSV-1	IFV-A	SARS-CoV2	HA-5	Reo-T1L
RLP068/I	++		+	+					++
RLP068/OTf	++		+	+	++	++	++	++	++
3Imid/I	++		+	+					
4Imid/I	++		+			+			–
4Imid/OTf	++		±	+					
4BuImid/OTf	++		±						
4DMAE/I	–		–			++			+
4DMAE/OTf	–		–			+			–
(Me3PhO)8Pc/I	–		–			+			++
8-4Imid/I	–		–			+			+

++ Strong activity (for bacteria this means reduction in CFUs to the limit of detection at 1 μM upon photoactivation with one or more light source; for viruses this means a > 99% reduction in viral activity at 10 μM). + Moderate activity (for bacteria this means reduction in CFUs to the limit of detection at 10 μM ; for viruses this means a 10–99% reduction in viral activity at 10 μM). ± Activity (for bacteria this means several logs of killing at 10 μM for one or more light source). – Minimal activity (for bacteria this means that no significant reduction in colony forming units is observed at $\leq 50 \mu\text{M}$; for viruses this means <10% reduction in viral activity). Blank cells were not determined.

concentrations (100 μM) **RLP068/OTf** is active toward *P. aeruginosa* even without light activation, causing a 6-log reduction in viable cells. PcMs **3Imid/I** and **4Imid/X** are less soluble than **RLP068/X** and thus were not tested at 100 μM . Other PcMs showed weaker efficacy, requiring 10–100 μM concentrations for a 1- to 3-log reduction in cell viability.

RLP068/OTf inactivates viruses in solution

RLP068/OTf was further tested for virucidal activity on a panel of pathogenic viruses representing all four physiochemical categories: influenza A virus (IFV-A) and SARS-coronavirus-2 (SARS-CoV-2), enveloped RNA viruses; herpes simplex virus type 1 (HSV-1), an enveloped DNA virus; human adenovirus 5 (HA-5), a non-enveloped DNA virus; and mammalian orthoreovirus Type I L (Reo-T1L), a non-enveloped RNA virus. Viruses were mixed with **RLP068/OTf** and exposed to light, then used to infect host cells and viral titers were determined. All viral titers were reduced to below the detection limit at 10 μM **RLP068/OTf** when photoactivated by all light sources tested, with the exception of Reo-T1L upon LED exposure (Table 1 and Fig. S3). These titers represent substantial reductions: IFV-A, HA-5 and SARS-CoV-2 infectivity were reduced ~ 3 logs and HSV-1 and Reo-T1L were reduced ~ 5 logs (LED resulted in a 3-log reduction of reo-T1L). The enveloped viruses HSV-1, IFV-A and SARS-CoV-2 also showed a 1- to 2-log reduction in titer with 1 μM **RLP068/OTf**, indicating greater sensitivity than the non-enveloped viruses to this PcM. No significant decrease in titer was observed for any of the viruses at 0.1 μM with any of the light sources.

Textile dyeing with highly active antimicrobial phthalocyanines

Having shown that low concentrations of the water-soluble PcMs **RLP068** and **Imid** have potent in-solution antimicrobial activity toward Gram-positive and Gram-negative bacteria and enveloped and non-enveloped viruses, these compounds were used to dye textiles to produce raw materials for PPE and other applications. The **RLP068/OTf** salt was used as it is more thermostable than **RLP068/I** (Fig. 1), allowing higher dyeing temperatures

(Fig. 2B), and **4Imid/I** was used as it is easier to synthesize than **3Imid/I**. Cotton, nylon and polyester were dyed with **RLP068/OTf** and **4Imid/I** under a range of concentrations, temperatures, times and washes to identify the minimal conditions resulting in coloured textiles that retained the dye upon washing and did not show visible leaching into water during overnight soaks. The dyeing parameters significantly impacted **RLP068/OTf** transfer from the dyeing solution to the substrates. Higher temperatures, longer dyeing times, and more concentrated dye solutions enhanced dye uptake. Once saturation was achieved, excess dye was released into the water upon soaking (*i.e.* visible leaching). As our goal is to achieve antimicrobial activity through direct contact of microbe with the dyed textiles, conditions were selected to avoid oversaturation and hence leaching. Though heating the solution shortens the dyeing time it adds an extra operational step, thus textiles were dyed at room temperature (rt). Solutions of **RLP068/OTf** with concentrations lower than 9 μM showed a noticeable decrease in colour intensity after dyeing a single textile sample, consistent with a reduction in concentration as the dye is absorbed onto the textile. A similar observation was made by Efimov *et al.* with the phthalocyanine dye LASU.¹² By increasing the **RLP068/OTf** concentration to 50 μM it was possible to recycle the dyeing solution at least 11 times, with a dyeing time of 90 seconds, without noticeable colour loss of the solution. This approach resulted in coloured cotton samples that did not exhibit detectable leaching after an overnight soak in water. **RLP068/OTf** was found to dye cotton well (Fig. 2A), whereas visible leaching occurred for nylon and polyester. **4Imid/I** dyes cotton well at rt and dyes cotton, nylon and polyester well at 80 °C (Fig. S4A) but visible leaching occurred for all three textiles. Furthermore, the higher temperature, 80 °C, caused **4Imid/I** solution to turn from deep blue-green to brown, suggesting degradation.

Cotton dyed with RLP068/OTf kills bacteria on contact with minimal leaching

To identify dyed fabrics that kill microbes on contact without substantial leaching into the surroundings we developed an



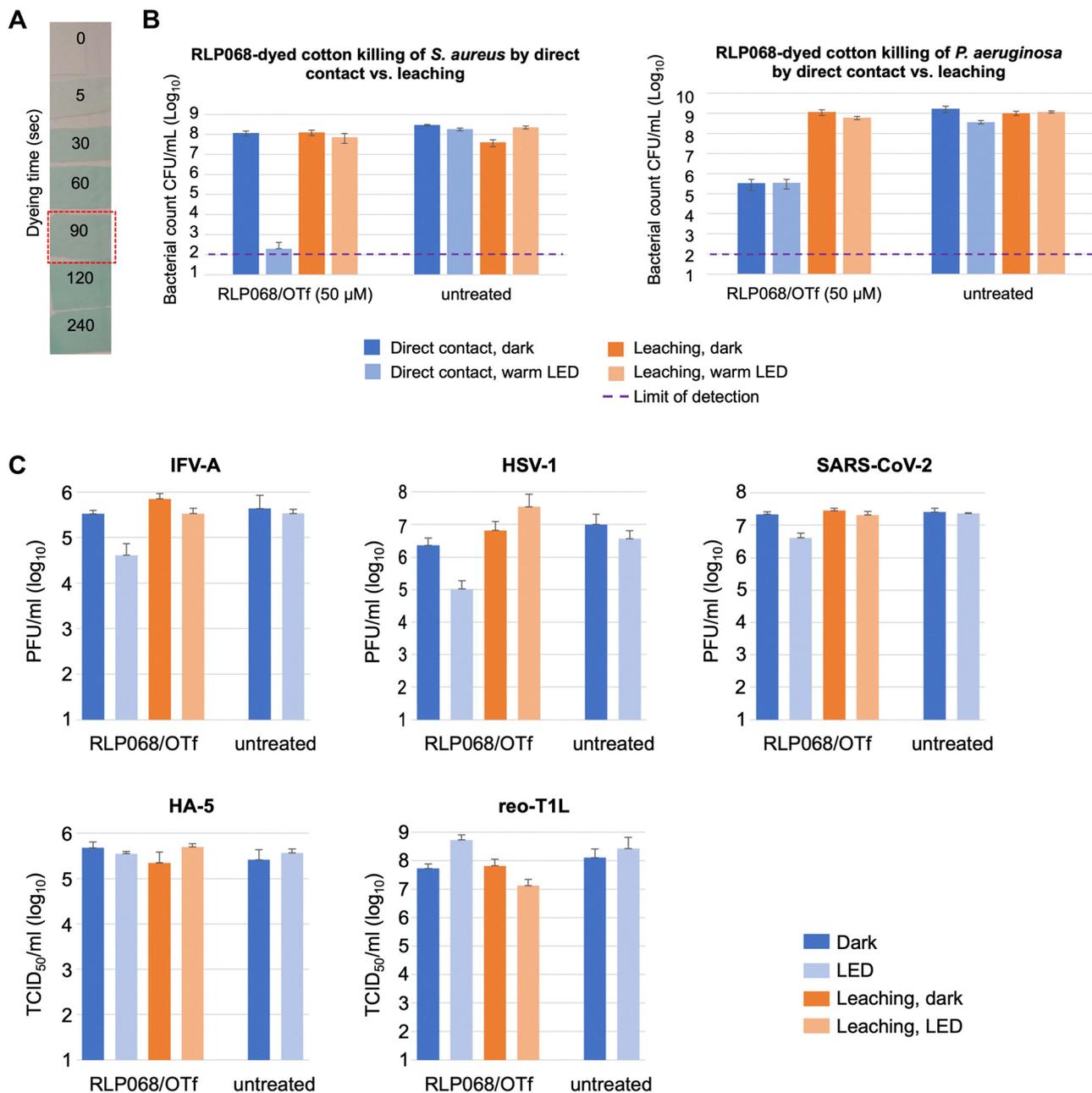


Fig. 2 Antimicrobial activity of PcM-dyed cotton. (A) Images of cotton samples dyed with 50 μM RLP068/OTf at rt for varying lengths of time. Cotton dyed with RLP068/OTf at rt for 90 s was tested for direct antimicrobial activity and leaching. (B) Antibacterial activity and leaching. Bacterial titers are shown as colony-forming units (CFUs). Standard error or the mean is shown for data from three experimental replicates. (C) Antiviral activity and leaching. Titters are shown as plaque-forming units (PFUs) or tissue culture infectious dose 50 (TCID₅₀). Untreated (undyed) cotton is shown as a negative control. Averages with standard deviation are shown for data from three experimental replicates.

assay that simultaneously tests, in a quantitative manner, (i) direct killing of bacteria dried onto the textiles and (ii) killing due to leaching of PcMs into the growth medium (Fig. S5). For the direct killing assay bacteria from overnight culture were applied to the dyed textile, dried for one hour to maximize contact between the cells and the textile, photoactivated with warm LED, and surviving cells were quantified. LED was selected as it is a common indoor light source and is more energy efficient and emits less heat than halogen. In parallel, a

leaching assay was performed on the dyed cotton by soaking it in phosphate-buffered saline (PBS) to allow the PcM to leach into the solution, and then testing the solution for antibacterial activity upon photoactivation. Direct killing/leaching assays were performed on dyed textiles that showed visible colour but did not show visible leaching into water after an overnight soak. Cotton dyed with 50 μM RLP068/OTf for 90 s at room temperature is a pale blue-green colour (Fig. 2A). This fabric showed high antibacterial activity, with a ~ 6 -log reduction in



colony forming units of *S. aureus* upon LED activation and a 3.5-log reduction in *P. aeruginosa* with and without photo-activation (Fig. 2B and Table S1). No antibacterial activity was detected for the solution in which the RLP068/OTf-dyed cotton had been soaked. As *S. aureus* exhibited several logs of killing in solution at 0.1 μM RLP068/OTf (Fig. S2A), we conclude that the RLP068/OTf concentration in the leaching solution is well below 0.1 μM , indicating minimal leaching.

Interestingly, despite the 4Imid/X PcMs showing robust killing of both Gram-negative and Gram-positive bacteria in solution (Fig. S2C-E and Table 1), and despite 4Imid/I dyeing cotton, nylon and polyester well, no antibacterial activity was observed for any of the 4Imid/I-dyed textiles (Fig. S4 and Table S1).

Cotton dyed with RLP068/OTf inactivates enveloped viruses

The virucidal activity of RLP068/OTf- and 4Imid/I-dyed cotton was tested with LED light exposure. Contact with RLP068/OTf-dyed cotton reduced the enveloped virus titers by 1–2-logs; approximately 90% of IFV-A and SARS-CoV-2 were inactivated, while 97% of HSV-1 was inactivated (Fig. 2C and Table S1). In contrast, no significant inactivation was observed for the non-enveloped viruses HA-5 and Reo-T1L. To test whether viral inactivation is due to direct contact with the dyed cotton or the leaching of RLP068/OTf into the medium, a leaching test was performed. The growth medium incubated with the dyed cotton did not exhibit virucidal activity. Thus, the antiviral effect of RLP068/OTf-dyed cotton on enveloped viruses appears to be a result of direct contact with the textile and not leaching, consistent with the results seen for bacteria. No antiviral activity was observed for 4Imid/I-dyed cotton, which is also consistent with the results for bacteria (Table S1).

Antibacterial efficacy of RLP068/OTf-dyed cotton is reduced with washing but not rubbing

To test the durability of the antibacterial effect of RLP068/OTf-dyed cotton under normal wear-and-tear conditions, samples were subjected to washing and rubbing. The dyed cotton was rinsed in water, followed by three-to-five 40-min wash cycles at 40 °C with or without rinsing to simulate the action of a regular washing machine. While a rinse step did not affect the dye intensity, washing with soap for 3 or more cycles removed most of the dye (Fig. 3A). Despite this fading the antibacterial activity for samples rinsed or washed with 3 cycles was retained at a level comparable to that of the unwashed sample, with 4–5 logs of killing. However, increasing the number of wash cycles to 5 resulted in a decrease in efficacy to \sim 2-logs killing, with further reduction to 1-log when two rinses are added to the wash cycles, demonstrating that the dye is not colourfast.

To assess the colour-fastness of the RLP068/OTf-dyed cotton upon application of friction, samples were placed on top of undyed cotton substrate and subjected to multiple cycles of rubbing, both by hand or using a motorized rubbing machine. Samples were inspected visually for transfer of dye from the RLP068/OTf-dyed cotton to the friction substrate. While machine rubbing visibly reduced the dye intensity of the RLP068/OTf-dyed cotton, it did not significantly impact its antibacterial activity (Fig. 3B). Transfer of the dye to the friction substrate was minimal as no colour was visible and no antibacterial activity was observed for this substrate. In summary, these results show that RLP068/OTf-dyed cotton retains its antibacterial efficacy upon rubbing and for at least 3 washes with soap but gradually loses efficacy with additional washes.

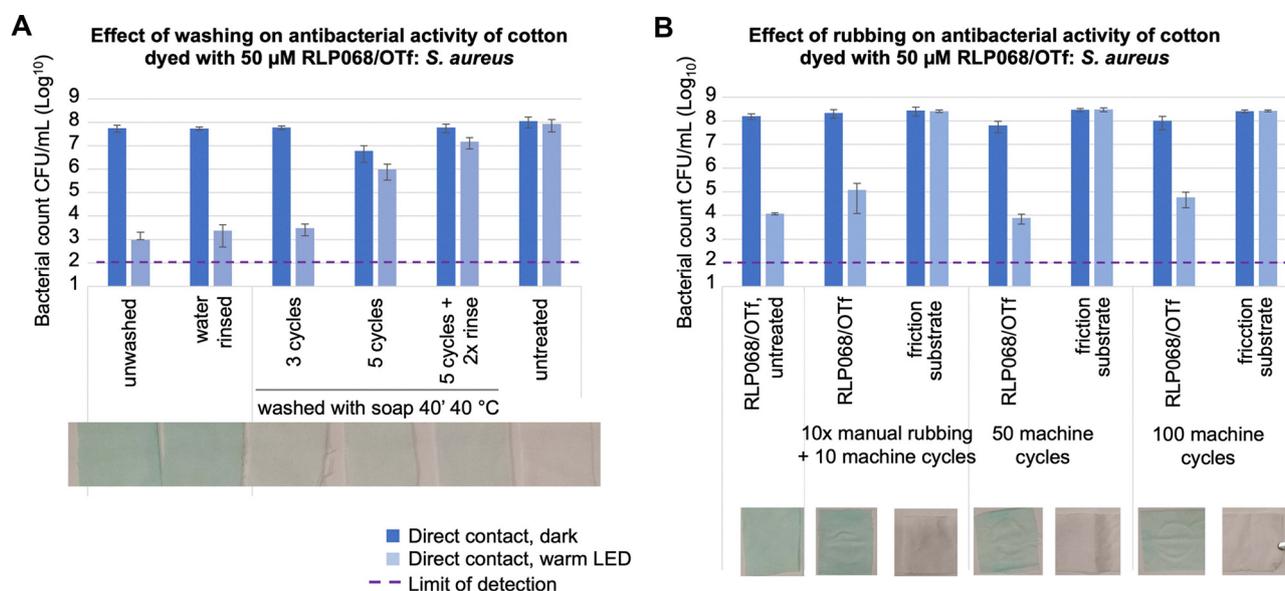


Fig. 3 Durability of antibacterial activity of RLP068/OTf-dyed cotton. (A) Cotton dyed with 50 μM RLP068/OTf (rt, 90 s) was rinsed or washed under various conditions and tested for activity against *S. aureus*. (B) RLP068-dyed cotton was rubbed against a friction substrate (untreated cotton) manually or in a rubbing machine and tested for activity against *S. aureus*. The friction substrate was also tested for transfer of antibacterial activity (PcM) from the dyed cotton. Untreated (undyed) cotton is shown as a negative control. Standard error of the mean is shown for data from three experimental replicates.



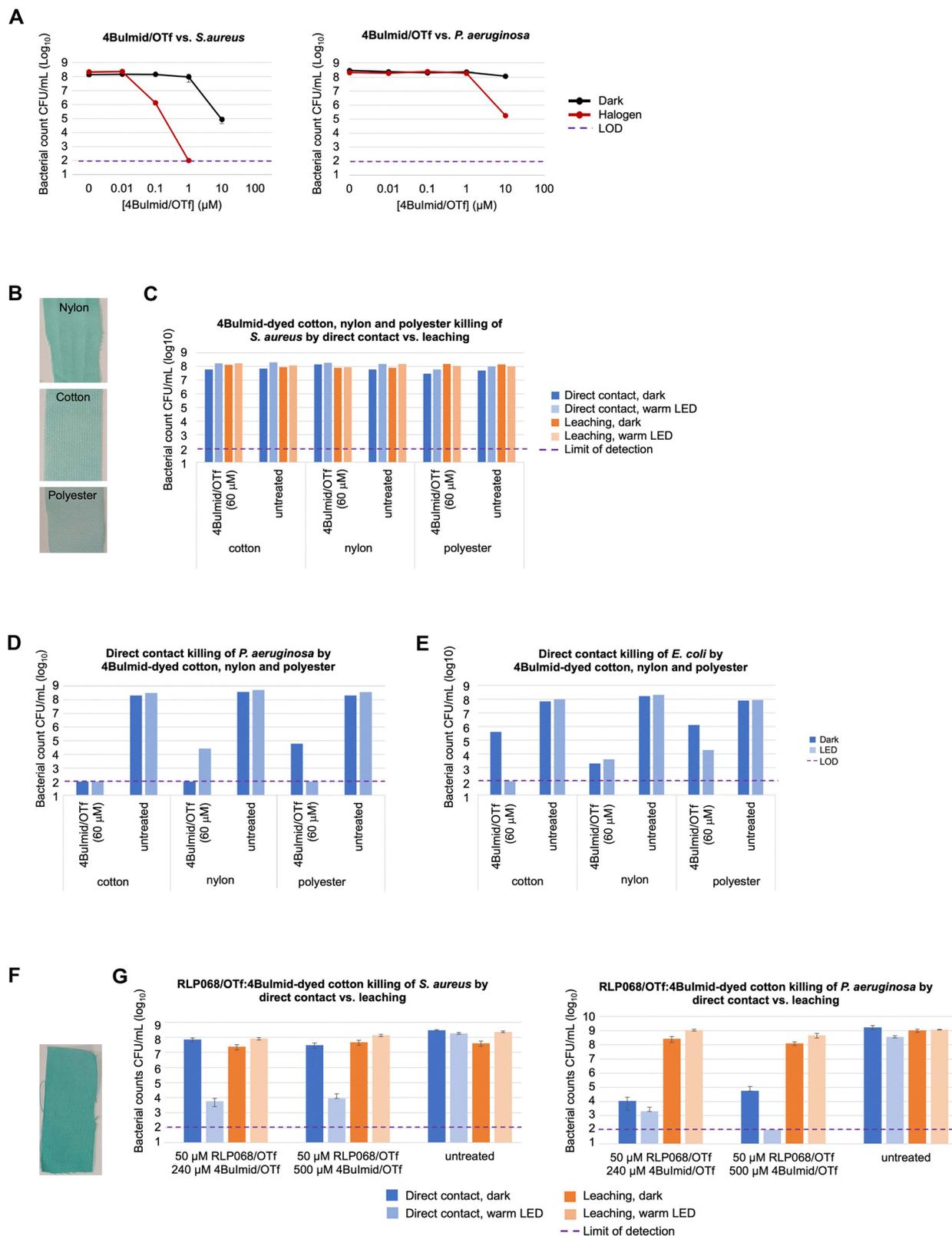


Fig. 4 Antibacterial activity of **4Bulmid/OTf** in solution and on textiles. (A) Antibacterial efficacy of **4Bulmid/OTf** in solution. (B) Images of cotton, nylon and polyester dyed with 60 μM **4Bulmid/OTf** for 60 min at 20 °C. Antibacterial activity of **4Bulmid/OTf**-dyed cotton, nylon and polyester toward (C) *S. aureus*, (D) *P. aeruginosa*, and (E) *E. coli* by direct contact. $N = 1$ for all. (F) Image of cotton dyed with a combination of 50 μM **RLP068/OTf** and 240 μM **4Bulmid/OTf**. (G) Antibacterial efficacy and leaching of cotton dyed with a combination of **RLP068/OTf** and **4Bulmid/OTf**. Untreated cotton is shown as a negative control. LOD, Limit of Detection. Standard error of the mean is shown for data from three experimental replicates.



Textiles dyed with a combination of water-soluble RLP068/OTf and water-insoluble 4BuImid/OTf show effective killing of both Gram-negative and Gram-positive bacteria

While RLP068/OTf-dyed cotton shows robust activity toward the Gram-positive bacterium *S. aureus*, it is less effective for Gram-negative bacteria (Fig. 2B). We thus produced and tested a water-insoluble PcM, 4BuImid/OTf, reasoning that this more hydrophobic molecule may interact with the hydrophobic outer membrane and thus be more effective at killing Gram-negative bacteria. Water-insoluble PcMs are also unlikely to leach into aqueous surroundings. Upon photoactivation with a halogen light, 4BuImid/OTf showed strong antibacterial activity against the Gram-positive *S. aureus* in solution, with an MIC of 1 μM , and significant activity (a 3-log reduction) at 10 μM with no light activation (Fig. 4A and Table S1). 4BuImid/OTf was less active than the water-soluble PcMs toward the Gram-negative *P. aeruginosa* in solution, showing only a 3-log reduction at 10 μM , compared to the 5–6-log reduction seen for RLP068/OTf, 3Imid/I, 4Imid/I and 4Imid/OTf at this concentration (Fig. S2).

Unlike RLP068/OTf, 4BuImid/OTf readily dyes the synthetic textiles nylon and polyester in addition to cotton, a natural fibre (Fig. 4B). Despite this, and despite the in-solution antibacterial activity of 4BuImid/OTf toward *S. aureus*, none of these dyed

materials showed any activity toward *S. aureus* (Fig. 4C). In contrast, all 4BuImid/OTf-dyed fabrics were highly active toward both *P. aeruginosa* and *E. coli*: both cotton and polyester produced a 6-log reduction in cell viability for *P. aeruginosa* and nylon reduced its viability by ~ 4 logs; cotton produced a 6-log reduction in *E. coli* and nylon and polyester produced 4–5-log reductions (Fig. 4D, E and Table S1). These dyed materials also show high potency in the absence of light activation, being almost as active as the photoactivated materials, and in the case of nylon, even more active than when photoactivated. These results imply that 4BuImid acts on Gram-negative bacteria via a mechanism that is independent of singlet oxygen, produced upon light activation of the PcMs. Leaching was tested using *S. aureus*, which is more sensitive than the Gram-negative bacteria to 4BuImid/OTf in solution. No leaching was observed for any of the textiles, as expected for this water-insoluble compound (Fig. 4C).

Since RLP068/OTf-dyed cotton is highly active against *S. aureus* (Fig. 2A) and 4BuImid/OTf-dyed cotton is highly active against *P. aeruginosa* and *E. coli* (Fig. 4D, E), cotton was dyed with a combination of these PcMs (Fig. 4F). Higher concentrations of 4BuImid/OTf were used in an effort to demonstrate activity toward *S. aureus*. This dye combination produced a

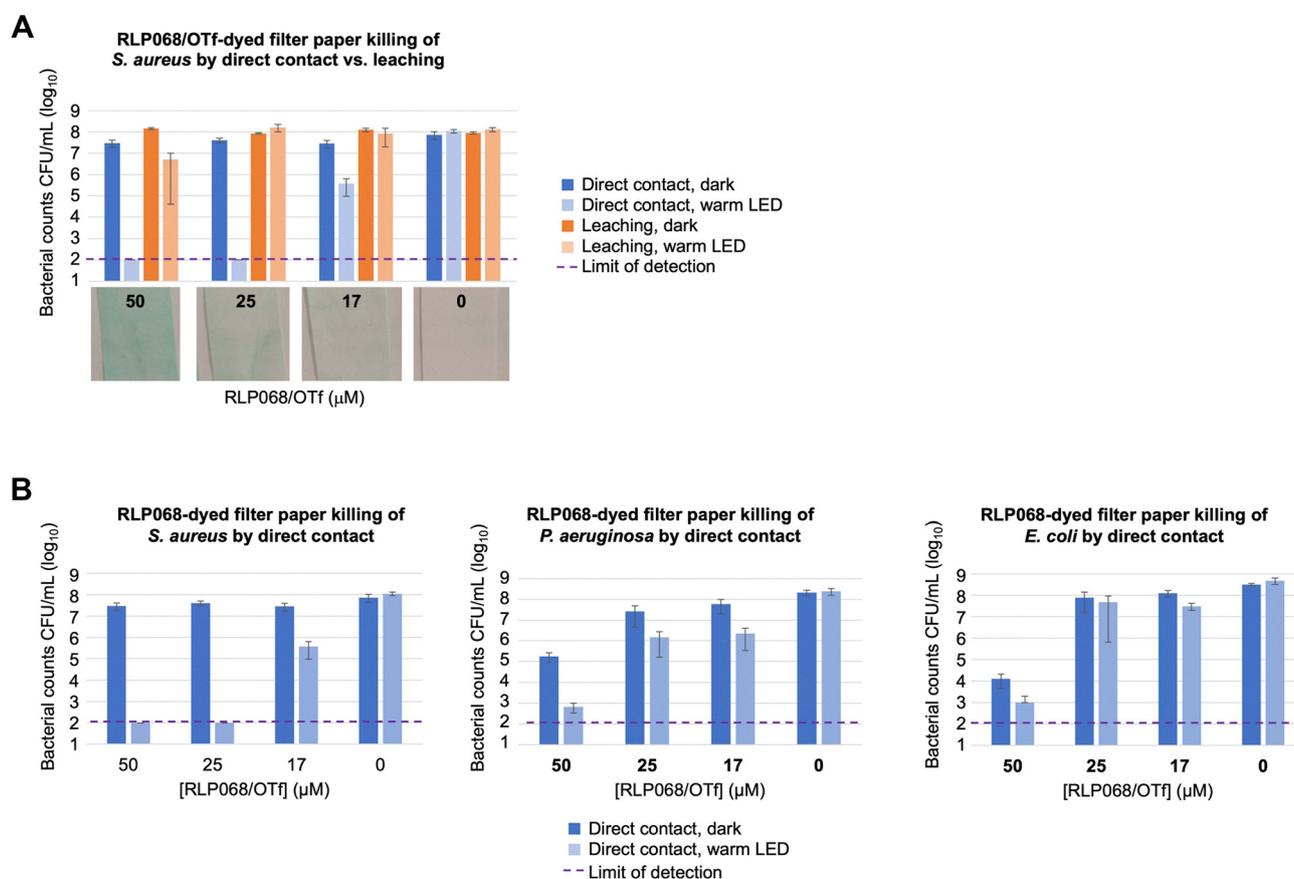


Fig. 5 Antibacterial activity and leaching results for RLP068-dyed paper. (A) Antibacterial efficacy and leaching of RLP068/OTf-dyed filter paper toward *S. aureus*. Images are shown of filter paper dyed with RLP068/OTf for 2 minutes at 20 $^{\circ}\text{C}$ at the indicated concentrations. (B) Antibacterial efficacy of RLP068/OTf-dyed filter paper toward *S. aureus*, *P. aeruginosa* and *E. coli*, assessed using a dry killing assay. Untreated filter paper is shown as a negative control. Standard error of the mean is shown for data from three experimental replicates.



~4-log reduction in *S. aureus* CFUs, which is less efficacious than **RLP068/OTf** alone. *P. aeruginosa* CFUs were reduced by 5–6 logs upon LED activation, and 3–4 logs without light (Fig. 4G). No leaching was detected for the combination dyes. Thus, the **RLP068/OTf/4BuImid/OTf** dye combination provides highly effective broad antibacterial activity.

RLP068-dyed paper has antibacterial activity

To test the potential use of photoactivatable PcMs in paper-based PPEs, filter paper was dyed with varying concentrations of **RLP068/OTf** and tested for antibacterial activity. The highest concentration of **RLP068/OTf**, 50 μM , produced a 6-log reduction in viable *S. aureus* but also leached into the medium, reducing CFUs by 1–2 logs; 25 μM produced a 6-log reduction with no leaching, and 17 μM reduced cell viability by only 2–3 logs without leaching (Fig. 5A). As observed for cotton, **RLP068/OTf**-dyed filter paper is less effective against Gram-negative bacteria, requiring 50 μM to reduce cell viability by 5 logs (Fig. 5B). Paper dyed with 25 μM **RLP068/OTf** showed only a 1-log reduction for the Gram-negative bacteria. These data show that paper dyed with low amounts of **RLP068/OTf** under mild conditions has significant self-sterilizing properties and thus has potential as a material for disposable PPEs.

Discussion

We demonstrate here the facile production of self-sterilizing materials, both cotton and paper, using **RLP068/OTf**, a thermostable salt of the **RLP068** cation, which is a known antimicrobial agent. We sought thermostable compounds that would withstand high temperatures potentially needed for dyeing and washing/drying. Though high temperatures (> 100 °C) were not required for the materials tested herein, they are required for dyeing synthetic materials. We identified optimal conditions of temperature, time and dye concentration that allow multiple samples to be dyed in the same solution, yielding textiles that are saturated with dye without leaching into the surroundings. The ability to dye textiles and paper under these mild conditions (room temperature, short duration, low dye concentration) in a cost-effective manner means that large-scale production can be undertaken to produce materials for longer lasting and more effective PPEs such as masks, caps and gowns.

Cotton dyed with **RLP068/OTf** provides excellent broad-spectrum antimicrobial activity: ~6-log reduction in viability of the Gram-positive bacterium *S. aureus*; a 3–4-log reduction in Gram-negative *P. aeruginosa*; and a 1–2-log reduction in activity for three enveloped viral pathogens. This activity matches or exceeds that reported for other dyed materials.^{12,21} Importantly, dyeing cotton with a combination of **RLP068/OTf** and the water-insoluble PcM **4BuImid/OTf** has robust activity toward both Gram-positive and Gram-negative bacteria. These textiles are promising candidates for PPEs.

Our in-solution and direct killing assays showed that Gram-negative bacteria, which have an outer membrane, are less susceptible to killing by singlet oxygen than Gram-positive

bacteria. The outer membrane of Gram-negative bacteria has a lipopolysaccharide outer leaflet that protects against antimicrobial agents including singlet oxygen.²⁶ The water-insoluble compound **4BuImid/OTf** showed weak activity toward the Gram-negative bacterium *P. aeruginosa* in solution yet is highly active toward both *P. aeruginosa* and *E. coli* on textiles, and this activity is largely light-independent. In contrast, cotton dyed with **4BuImid/OTf** shows no activity toward the Gram-positive *S. aureus*. We hypothesize that **4BuImid/OTf** kills Gram-negative bacteria by interacting with and disrupting the outer membrane rather than through singlet oxygen production. This effect may be more apparent with the dyed textiles, which allow the bacteria, condensed in a dried drop, to intimately contact the densely distributed PcM, receiving a higher effective dose than they would in solution. The combination of **RLP068/OTf** and **4BuImid/OTf** on cotton is effective for both Gram-positive and Gram-negative bacteria, with **RLP068/OTf** acting on both in a light-dependent manner and **4BuImid/OTf** acting on the *P. aeruginosa* outer membrane in a light-independent manner.

The virucidal activity of the PcMs was less dramatic than the bactericidal activity. Furthermore, while all virus types were inactivated by several logs by **RLP068/OTf** in solution, only the enveloped viruses were inactivated on dyed cotton, and with considerably less efficiency. These results may reflect the design of the experiment to test the antiviral activity of **RLP068/OTf**-dyed cotton, in which the virus was applied in solution rather than dried onto the fabric, as was done for bacteria. The drying step was not performed because viruses, especially enveloped ones, are sensitive to dehydration. Indeed, Efimov *et al.* demonstrated significant virucidal activity of HCoV-229E on photoactivated phthalocyanine-dyed cotton after drying the virus sample on the cotton, but also saw a 2-log reduction by drying alone.¹² In our experimental setting, viruses in solution must collide with singlet oxygen produced at the fabric surface. Singlet oxygen diffuses ~100 nm²⁷ in solution and has a lifetime of ~4 μs ,²⁸ and these values will be reduced by the presence of proteins in the medium.²⁹ In the environment, viruses within respiratory droplets that settle on PcM-treated PPE would be inactivated by both dehydration and oxidation from singlet oxygen. The difference in PcM susceptibility of enveloped viruses vs. non-enveloped viruses has been observed for other disinfectants.^{30,31} Singlet oxygen oxidizes unsaturated lipids in the viral envelope, changing its biophysical properties and disrupting viral membrane fusion to host cells.³² As the viral envelope is a single continuous unit, damage at one or a few sites can disrupt its integrity, inactivating the virus. In contrast, singlet oxygen-induced damage to one or more proteins on a non-enveloped viral capsid would not necessarily disrupt the entire capsid and may thus require higher numbers of singlet oxygen collisions. Though not tested here, enveloped viruses are expected to be susceptible to **4BuImid/OTf**, which may disrupt their envelope as it appears to do for the outer membrane of Gram-negative bacteria.

Dyeing textiles with PcMs is a low-cost alternative to covalent attachment or application of antimicrobial coatings. The PcMs described in this study can be synthesized in large volumes and



the dyeing process requires very basic infrastructure (e.g. vats), as textiles are simply dipped or soaked for a short time in the dye then washed in water and air-dried, all at room temperature. In contrast, covalent attachment of antimicrobial compounds requires more complex synthetic efforts and conditions, and nanoparticles, copper and other metals are relatively costly and can require special handling.

Conclusion

In summary, our results show that broad-spectrum antimicrobial textiles active against bacteria and viruses can be produced inexpensively for use in disposable PPE such as masks, caps and cloths, where the (photo)active antimicrobial phthalocyanine-based complexes are non-covalently bound to the fabric. These PPE can be used for longer periods of time than untreated PPE, which will help to address supply chain issues, and will provide greater protection, limiting microbial transmission.

Experimental

Synthesis and characterization of metallophthalocyanines

Synthetic procedures and characterization details for new compounds are presented in the SI, along with general methods and instrumentation details. 3-(3-(dimethylamino)phenoxy)phthalonitrile, $(\text{Me}_2\text{NPhO})_4\text{PcZn}$, **RLP068/I**, and $(\text{Me}_3\text{N-PhO})_8\text{PcZn/I}$ were synthesized using the reported procedures.¹⁵ $(\text{Me}_2\text{NPhO})_4\text{PcZn}$ was cyclized and metalated in one step from the respective phthalonitrile and zinc acetate, and **RLP068/I** was prepared *via* the reaction of iodomethane and $(\text{Me}_2\text{NPhO})_4\text{PcZn}$. **4Imid**,²⁵ **4DMAE/I**³³ and **8-4Imid/I**²⁴ were prepared as previously reported.

Preparation of textiles

The textiles used for dyeing with PcMs were cotton (100%, 115 g m⁻²), nylon (100%, 200 g m⁻²), and polyester (100%, 163 g m⁻²) from Fabricana Ltd. Prior to dyeing, samples of each material (12.5 g) were cleaned in 1.4 L of washing solution containing 2% Synthrapol and 4.4% sodium carbonate (w/v) while stirring at 100 rpm for 60 min at 80 °C. After cleaning, the textiles were rinsed with running tap water for 5 min, soaked in purified water (> 5 MΩ cm, Millipore Elix 100) for 60 min, and dried for at least 16 h. Paper samples were dyed using filter paper (Grade 1, pore size 11 μm, GE Healthcare) as substrate. Dyeing solutions and dyed fabrics were stored in the dark to prevent degradation. In a standard dyeing procedure,³⁴ samples were cut into strips of a predetermined weight (cotton, 100 mg; nylon, 120 mg; polyester, 130 mg; paper 600 mg) and dyed by immersion in the PcM solutions in 20 mL glass vials. Dyeing conditions, including PcM concentrations, are reported in Table S1. Samples dyed with **RLP068/OTf** or **4Imid/I** were removed from the vials and air-dried in the dark for at least 16 h. Samples dyed with **4BuImid/OTf** dissolved in dimethyl sulfoxide (DMSO) were placed in a fume hood overnight to evaporate the DMSO. Samples were thoroughly rinsed with tap

water and soaked in purified water for 10 min. For the **RLP068/OTf** and **4BuImid/OTf** combination dye, the previously rinsed and dried **4BuImid/OTf** samples were immersed in 50 μM **RLP068/OTf** for 90 s at 20 °C, dried for at least 16 h and stored in the dark.

Testing the effect of washing on retention of antibacterial activity for dyed fabrics

The durability of the antibacterial activity of **RLP068/OTf**-dyed cotton upon rinsing or washing was evaluated. Dried cotton samples dyed with 50 μM **RLP068/OTf** (90 s, 20 °C) were left untreated or subjected to a rinse cycle (rinsed with 2 L of flowing tap water, soaked in 1 L of purified water for 10 min, then dried for a minimum of 16 h). Some samples were further subjected to one or more wash cycles where they were stirred at 400 rpm for 40 min in 1 L of tap water containing laundry detergent (Persil, 0.8 wt%) at 40 °C. Following washing, samples were rinsed with tap water followed by purified water to remove any residual detergent. Some samples were further soaked in purified water for 40 minutes twice and then air-dried overnight. Colour-fastness was assessed visually and using the dry-killing assay to test for antibacterial activity.

Testing the effect of rubbing/friction on retention and transfer of antibacterial activity for dyed fabrics

The durability of the antibacterial activity of **RLP068/OTf**-dyed cotton when subjected to rubbing (crocking) was tested manually or on a motorized rubbing machine (Crocktec, Roaches International) using the American Association of Textile Chemists and Colorists (AATCC) test method 8,³⁵ with untreated (undyed) cotton used as the friction rubbing substrate. For machine rubbing the dyed cotton was fixed with a metal clip to a 16 mm rubbing head attached to a mechanical arm, which applies a 9 N force to the friction substrate fixed to a mounting plate. One rubbing cycle consisted of two rubbing strokes of 104 mm. Manual rubbing involved the same procedure except the cycles were done by hand. Samples were subjected to 20 (10 manual cycles plus 10 machine cycles), 50 (machine) and 100 (machine) rubbing cycles. The retention of dye on the **RLP068**-treated cotton and its transfer to the undyed cotton upon rubbing were assessed visually and by dry-killing assay.

Bacterial and viral strains and host cells

Bacteria. Methicillin-resistant *S. aureus* (ATCC 43300) was purchased from the American Tissue Culture Collection (ATCC). *P. aeruginosa* (strain K) and *E. coli* (DH5α) were from in-house stocks.

Viruses. HSV-1 F strain with a deletion in thymidine kinase was recovered by transfecting an infectious BAC clone, pYE-bac102 (a gift from Y. Kawaguchi) to Vero cells.³⁶ IFV-A (A/California/07/09 pH1N1, NR-13663) and SARS-CoV-2 (Isolate USA-WA1/2020, NR-52281) were obtained from BEI Resources. HA-5 (Adenoid 75, VR-5) and Reo-T1L (Lang1, VR-230) were purchased from ATCC. MDCK (ATCC CCL-34), Vero (ATCC CCL-81), HeLa (ATCC CCL-2) and L929 (ATCC CCL-1) cell lines were obtained from ATCC, while VeroE6-TMPRSS2 (JCRB1819)



was from the JCRB Cell Bank. Cells were maintained in Dulbecco's modified Eagle medium (DMEM, Sigma) supplemented with 10% fetal bovine serum (FBS) and antibiotics (100 IU penicillin and 100 $\mu\text{g mL}^{-1}$ streptomycin) except for L929, which was maintained in minimum essential medium (MEM, Sigma) with the same supplements. Cells were grown at 37 °C in 5% CO₂. The experiments involving SARS-CoV-2 were performed in BIO3, a containment level 3 laboratory at SFU Burnaby campus as approved by the SFU biosafety committee (permit #361-2021, 2022 and 2023).

In-solution antibacterial activity assay

Methicillin-resistant *S. aureus* and *P. aeruginosa* were grown in Lysogeny Broth (LB) overnight at 37 °C, shaking at 250 rpm. Overnight cultures were diluted 1:100 in LB and grown to OD₆₀₀ 0.5. Cells (1 mL) were washed twice with PBS by centrifuging at 3000 $\times g$ for 10 min followed by resuspension in 1 mL PBS. To prevent background photoactivation of the PcMs all subsequent steps were performed with minimal light unless indicated. Ten μL of resuspended cells was added to 90 μL PcM at various concentrations in 96 well plates. The plates were incubated at room temperature (approx. 20 °C) in the dark or illuminated from above for 1 h at 3000 lux ($\pm 3\%$), measured using the AP-881D Digital Lux Meter from AOPUTTRIVER. The light sources used were: a halogen Heath Zenith Security Light, SL-5505 (500 W, 6.0 Hz, 4.2 A); a T8 warm LED tube light (2 foot, 24 W, AC100–305 V 50/60 Hz, 3000–3500 K Dong Guan City Shengwei Lighting Co.); and a Sylvania T12 fluorescent light tube, SL21552 (18 inches, 15 W, 3000 K). Samples were serially diluted and 10 μL aliquots were plated on LB agar. Plates were incubated in the dark at 37 °C for 18 h and the number of colony-forming units (CFUs) was determined for each PcM and condition as well as for untreated controls. Experiments were performed in triplicate except where indicated.

Dry killing antibacterial activity assay

Bacteria were prepared as for in-solution bactericidal assays. Washed cells were resuspended in PBS with 10% LB. All steps were carried out under minimum light conditions unless indicated. Dyed and untreated textiles were cut into 7 \times 7 mm squares and placed in wells of a 24-well plate. A 20 μL aliquot of resuspended cells was applied evenly to the textile and air dried for 1.5 h in a biosafety cabinet. Plates were incubated at room temperature in the dark or illuminated from above at 3000 lux for 1 h. Cells were resuspended in 200 μL LB by gentle pipetting then serially diluted, plated and colonies were counted as described above. Experiments were performed in triplicate except where indicated.

Leaching assay for bacteria

The antibacterial activity of the growth medium in which PcM-treated textiles were soaked was tested to detect leaching of the PcMs. Dyed and untreated textiles were cut into 7 \times 7 mm squares and placed in wells of a 48-well plate. Twenty μL of PBS was added to each well and the plates were incubated at room temperature for 1 h. An additional 20 μL PBS was added and mixed, and 10 μL was transferred into wells of 96-well plates, to

which 10 μL of washed cells was added. Plates were left in the dark or illuminated from above at 3000 lux for 1 h. Cells were serially diluted, plated and colonies were counted as described above.

In-solution antiviral activity assay

To test the antiviral activity of the solution PcMs, compounds were dissolved initially in DMSO and then diluted at least 100-fold in DMEM with antibiotics and mixed with the test virus stock solution in 96-well plates (20 μL per well) in triplicate. Solutions were mixed in an unlit biosafety cabinet to minimize light exposure. The plates containing the mixtures were exposed for 1 h to warm LED, fluorescent, or halogen light by the same devices under the same conditions as for the antibacterial activity assay or kept in the dark. The light exposure was performed in a darkroom to minimize ambient light; samples were exposed to halogen light in a dark 4 °C room to avoid heating the samples. The light-exposed samples were pooled and stored at -80 °C in aliquots until titrated.

On-textile antiviral activity assay

To test the antiviral activity of RLP068-dyed cotton, 6 mm circles of the fabric were placed in triplicate wells of a 96-well plate and overlaid with 20 μL per well of virus stock. The plate was exposed to LED light as described above. After the light exposure, DMEM (180 μL) was added to the well and the entire volume was recovered and stored at -80 °C until the titration. To determine if virucidal activity is due to direct contact between the viruses and the dyed cotton or to leaching of RLP068/X into the solution, a leaching assay was performed. The dyed cotton was soaked for 1 h in 20 μL of DMEM in a 96-well plate in the dark, then 20 μL of this soaking solution was mixed with 20 μL of the virus stock in a new 96-well plate and exposed to the LED light for 1 h. This sample was stored at -80 °C prior to titration.

Virus titration

HSV-1 titers were determined by a plaque assay on Vero cells. Vero cell monolayers in 12-well plates were inoculated with 100 μL of HSV-1 samples. After 1 h adsorption, the monolayers were overlaid by DMEM containing 1% methylcellulose and 2% FBS. The plates were incubated at 37 °C for 3 days and the plaque forming units (PFUs) were counted after staining cells with 0.5% crystal violet in 20% methanol. SARS-CoV-2 titration was performed with VeroE6-TMPRSS2 cells in 12-well plates in the same manner as for HSV-1, except that the monolayers were fixed by methanol:acetone (1:1) before staining. IFV-A titration was performed with MDCK cells in 6-well plates with 200 μL virus sample inoculum. The inoculated monolayers were overlaid with DMEM containing 1% agar noble and 0.00075% trypsin (w/v). Plates were incubated at 37 °C for 4 days and cells were stained with neutral red to count PFUs. Reo-T1L and HA-5 were titrated by tissue culture infectious dose 50 (TCID₅₀) in 96-well plates. For Reo-T1L, a 10-fold serial dilution of the viral sample was prepared and each dilution was mixed with L929 cells (1 \times 10⁴/200 μL) in MEM with 5% FBS. The mixtures were



dispensed to a 96-well plate (200 μ L per well, 12 wells per dilution). The infection in each well was determined after 5 day incubation by visible cytopathic effect (CPE) under a light microscope. The HA-5 titer was determined similarly to Reo-T1L but with HeLa cells in DMEM with 10% FBS. After 5 days of incubation, the cells in 96-well plates were fixed by a 1 : 1 mixture of methanol and acetone and cells were stained using an HA-5 hexon-specific monoclonal antibody (TC31-9C2.C9, Development Studies Hybridoma Bank), horse radish peroxidase-labeled anti-mouse IgG (Jackson Laboratory), and the enzymatic substrate 3,3'-diaminobenzidine (Vector). Positively stained wells were counted as infection-positive wells. The TCID₅₀ was calculated using the Reed and Muench method.

Conflicts of interest

There are no conflicts to declare.

Data availability

The authors declare that all data supporting the findings of this study are available within the article and its supplementary information (SI) files. Supplementary information: additional synthetic and characterization details for phthalonitriles and metallophthalocyanines and other experimental details; antimicrobial activity data for PcM in solution and PcM-dyed textiles. See DOI: <https://doi.org/10.1039/d5ma00607d>.

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References

- 1 FAO, UNEP, WHO and WOA, *One Health Joint Plan of Action, 2022–2026*, Rome, Italy, 2022.
- 2 W. A. Rutala and D. J. Weber, *Guideline for Disinfection and Sterilization in Healthcare Facilities*, 2008.
- 3 P. Wang, W. D. Cutts, H. Ning, S. Pillay and S. Liu, *J. Polym. Res.*, 2022, **29**, 360.
- 4 S. Paul, M. Dhangar, K. Chaturvedi, H. Bajpai, N. Siraj, R. K. Mohapatra, S. Sundaramurthy, S. Arisutha, B. Jethy, B. Naik, M. A. Khan, A. K. Srivastava and S. Verma, in *Advanced Materials and Conversion Technologies for Personal Protective Equipment Used in the COVID-19 Pandemic*, ed. S. Sundaramurthy, S. Verma and A. K. Srivastava, Springer Nature, Singapore, 2024, pp. 151–178.
- 5 A. Purniawan, M. I. Lusida, R. W. Pujiyanto, A. M. Natri, A. A. Permasari, A. A. H. Harsono, N. H. Oktavia, S. T. Wicaksono, J. R. Dewantari, R. R. Prasetya, K. Rahardjo, M. Nishimura, Y. Mori and K. Shimizu, *Sci. Rep.*, 2022, **12**, 4835.
- 6 B. S. T. Peddinti, F. Scholle, R. A. Ghiladi and R. J. Spontak, *ACS Appl. Mater. Interfaces*, 2018, **10**, 25955–25959.
- 7 S. R. D. Gamelas, C. Vieira, M. Bartolomeu, M. A. F. Faustino, J. P. C. Tomé, A. C. Tomé, A. Almeida and L. M. O. Lourenço, *J. Photochem. Photobiol., B*, 2022, **233**, 112502.
- 8 N. Kaushik, S. Mitra, E. J. Baek, L. N. Nguyen, P. Bhartiya, J. H. Kim, E. H. Choi and N. K. Kaushik, *J. Adv. Res.*, 2023, **43**, 59–71.
- 9 K. A. Mariewskaya, A. P. Tyurin, A. A. Chistov, V. A. Korshun, V. A. Alferova and A. V. Ustinov, *Molecules*, 2021, **26**, 3971.
- 10 A. Mathur, A. S. Parihar, S. Modi and A. Kalra, *Microb. Pathog.*, 2023, **183**, 106307.
- 11 F. Vigant, N. C. Santos and B. Lee, *Nat. Rev. Microbiol.*, 2015, **13**, 426–437.
- 12 A. Efimov, C. Dagallier, C. Frochot, B. Myrzakhmetov, P. Arnoux, T. Heinonen, M. Mannerström, T. Toimela, Z. Ahmed, J. F. Audibert, B. Habermeyer, S. Mordon and R. B. Pansu, *Photodiagn. Photodyn. Ther.*, 2024, **45**, 103978.
- 13 A. Almeida, A. Cunha, M. A. F. Faustino, A. C. Tomé and M. G. P. M. S. Neves, in *Photodynamic Inactivation of Microbial Pathogens: Medical and Environmental Applications*, ed. M. R. Hamblin and G. Jori, The Royal Society of Chemistry, 2011, vol. 11, pp. 83–160.
- 14 A. Galstyan, *Chem. – Eur. J.*, 2021, **27**, 1903–1920.
- 15 D. Dei, G. Chiti, M. P. De Filippis, L. Fantetti, F. Giuliani, F. Giuntini, M. Soncin, G. Jori and G. Roncucci, *J. Porphyr. Phthalocyanines*, 2006, **10**, 147–159.
- 16 P. S. da Silva Santos, B. da Fonseca Orcina, R. R. G. Machado, F. V. Vilhena, L. M. da Costa Alves, M. S. R. Zangrando, R. C. de Oliveira, M. Q. S. Soares, A. N. C. Simão, E. C. I. N. Pietro, J. P. G. Kuroda, I. A. de Almeida Benjamim, D. B. Araujo, S. H. Toma, L. Flor, K. Araki and E. L. Durigon, *Sci. Rep.*, 2021, **11**, 19937.
- 17 Z. Smetana, E. Mendelson, J. Manor, J. E. van Lier, E. Ben-Hur, S. Salzberg and Z. Malik, *J. Photochem. Photobiol., B*, 1994, **22**, 37–43.
- 18 N. E. Grammatikova, L. George, Z. Ahmed, N. R. Candeias, N. A. Durandin and A. Efimov, *J. Mater. Chem. B*, 2019, **7**, 4379–4384.
- 19 T. Wright, M. Vlok, T. Shapira, A. D. Olmstead, F. Jean and M. O. Wolf, *ACS Appl. Mater. Interfaces*, 2022, **14**, 49–56.
- 20 T. J. Cuthbert, S. Ennis, S. F. Musolino, H. L. Buckley, M. Niikura, J. E. Wulff and C. Menon, *Sci. Rep.*, 2021, **11**, 19029.
- 21 L. George, A. Hiltunen, V. Santala and A. Efimov, *J. Inorg. Biochem.*, 2018, **183**, 94–100.
- 22 R. T. Aroso, M. J. F. Calvete, B. Pucelik, G. Dubin, L. G. Arnaut, M. M. Pereira and J. M. Dąbrowski, *Eur. J. Med. Chem.*, 2019, **184**, 111740.
- 23 C. P. S. Ribeiro and L. M. O. Lourenço, *J. Photochem. Photobiol., C*, 2021, **48**, 100422.
- 24 V. Lioret, S. Saou, A. Berrou, L. Lernerman, C. Arnould and R. A. Decréau, *Photochem. Photobiol. Sci.*, 2023, **22**, 303–309.
- 25 X. F. Zhang, Y. Lin, W. Guo and J. Zhu, *Spectrochim. Acta, Part A*, 2014, **133**, 752–758.
- 26 T. A. Dahl, R. W. Midden and P. E. Hartman, *Photochem. Photobiol.*, 1987, **46**, 345–352.
- 27 R. W. Redmond and I. E. Kochevar, *Photochem. Photobiol.*, 2006, **82**, 1–178.



- 28 A. A. Krasnovsky, *Biochemistry*, 2007, **72**, 1065–1080.
- 29 Ł. Ożog and D. Aebisher, *Eur. J. Clin. Exp. Med.*, 2018, **16**, 123–126.
- 30 P. Tarka and A. Nitsch-Osuch, *Viruses*, 2021, **13**, 534.
- 31 L. Zeng, J. Li, M. Lv, Z. Li, L. Yao, J. Gao, Q. Wu, Z. Wang, X. Yang, G. Tang, G. Qu and G. Jiang, *Environ. Health*, 2023, **1**, 15–31.
- 32 L. Zeng, M. Di Wang, S. L. Ming, G. L. Li, P. W. Yu, Y. L. Qi, D. W. Jiang, G. Y. Yang, J. Wang and B. B. Chu, *Redox Biol.*, 2020, **36**, 101601.
- 33 D. Wöhrle, N. Iskander, G. Grasczew, H. Sinn, E. A. Friedrich, W. Maier-Borst, J. Stern and P. Schlag, *Photochem. Photobiol.*, 1990, **51**, 351–356.
- 34 A. K. R. Choudhury, *Handbook of Textile and Industrial Dyeing*, ed M. Clark, 2011, **1**, 64–149.
- 35 AATCC, *Test Method For Colorfastness To Crocking: Crockmeter: AATCC TM8-2016e*, 2022.
- 36 M. Tanaka, H. Kagawa, Y. Yamanashi, T. Sata and Y. Kawaguchi, *J. Virol.*, 2003, **77**, 1382–1391.

