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Photonic Designs to Reduce Morbidity and Mortality for ICU Patients on Ventilators

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Photonic designs to reduce morbidity and mortality for ICU patients on ventilators

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ABSTRACT

Ventilator-associated pneumonia (VAP) is pneumonia that occurs >48hrs after initiation of mechanical ventilation and is a significant cause of morbidity and mortality in patients that are hospitalized in intensive care units (ICU). The risk of developing VAP increases during use, and a diagnosis of VAP has been associated with a substantial cost. There are up to hundreds of thousands of cases in the US per year, costing the healthcare system billions annually. Patients who suffer from VAP frequently require longer ICU stays, higher exposure to antibiotics, and more hospital care at the risk of increased mortality. The SARS-CoV-2 pandemic has further increased the use of antibiotics among patients with COVID-19, an indicator of increased VAP prevalence. Before 2020, strides were made to reduce the incidence of VAP through hygienic protocols known as 'VAP bundles.' Despite the improvements, VAP continues to be a large problem, with the inoculation of pathogens within the endotracheal tube (ETT) itself. ETTs with built-in subglottic suction devices (SSD-ETT) allow the removal of subglottic secretions, but this has been adopted heterogeneously. We propose novel optical device designs to be used in combination with SSD-ETTs to reduce colonization and biofilm formation on the inner lumen of ETTs and reduce the incidence of VAP and improve patient care.

Keywords: Ventilator-associated pneumonia (VAP), intensive care units (ICU), SARS-CoV-2 pandemic, endotracheal, biofilm, infection, ultraviolet light (UV)

1. INTRODUCTION

The incidence of ventilator-associated pneumonia (VAP) increases continually > 48 hours beyond the initial intubation of mechanical ventilation at 2 to 16 per 1000 ventilator days, with 10% of patients in the ICU developing VAP [1,2]. VAP substantially impacts the morbidity and mortality of patients hospitalized in intensive care units (ICU), with a 10-30% mortality risk[1]. The United States healthcare system responds to over 300,000 cases of VAP, costing $2.6-3.6 billion annually, with additional care costs equating to $40,000 per patient[2,3]. VAP is associated with longer durations of mechanical ventilation and ICU stays, increased exposure to broad-spectrum antibiotics, higher requirements for staffing and patient care, and increased mortality[2]. The SARS-CoV-2 pandemic has put an additional strain on the healthcare system, especially in ICUs. While data on VAP infections are currently inadequate, reported antibiotic use among patients hospitalized with COVID-19 infection is estimated at between 71-100%, indicating VAP bacterial infections are a major risk to COVID-19 patients[4].

Point of care improvements in recent years, collectively known as a 'VAP bundle,' have reduced the incidence of VAP through hygienic engineering and protocol alterations[5]. Despite this, VAP continues to be a large problem, with the leading cause being the micro-aspiration of subglottic secretions and biofilm formation on the inside and outside of the endotracheal tube (ETT). The structured, multicellular, differentiated, and syntrophic bacterial communities of biofilms pose a large threat to hospitals due to their resistance and adaptability to a variety of antimicrobial agents[6]. To combat the buildup of potentially infectious secretions, ETTs engineered for VAP bundle use include built-in subglottic suction devices (SSD-ETT) to remove subglottic secretions[7]. This point of care addition is recommended by the CDC and American Thoracic Society for patients expected to require greater than 48-72 hours of mechanical ventilation, but this has been adopted heterogeneously[8].

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The formation of biofilms on medical devices poses a large threat to the well-being of patients, with biofilms present in 65% of all hospital infections and found in 95% of all ETTs post-intubation[6,9]. The presence of biofilms is strongly linked to increases of risk factors in morbidity and mortality[6,10]. Recent studies have sought to limit or restrict the formation of biofilms early in their adhesion with targeted ultraviolet light, preventing biological material from seeding pathogenic organisms[10]. UV light in the 280-100 nm region (UVC) is known to have multiple modes of biological inhibitory responses within seconds of illumination, rapidly and drastically reducing biofilms[10,11]. The inclusion of a novel, rapid-sterilizing UVC catheter system into the VAP bundle could reduce the incidence of biofilm formation on the inner lumen of ETTs and resultantly, VAP cases themselves.

Suitable surrogate bacteria were required to study the germicidal effects of UVC. VAP-related organisms are most frequently associated with Gram-negative biofilm-forming bacteria, contributing toward 60% of all cases, with the remaining being largely composed of Gram-positive bacteria[12]. The family of Enterobacteriaceae is responsible for 14.1% of all cases of VAP, with hospital-derived *Escherichia coli* (*E.* coli) being one of the most prominent sources of pathogenesis[12]. Subset from Enterobacteriaceae, the *E. coli* phylogroup B2 is most frequently associated with VAP[13,14]. The model organism *Escherichia coli* (ATCC® 25922™) was selected as a surrogate pathogenic organism model for Gram-negative biofilm-forming bacteria due to its location within phylogroup B2, its pathogenic biofilm formation behaviors, and its use in prior UV sterilization studies[15-18]. To account for the Gram-positive biofilm-forming bacteria, *Bacillus subtilis subsp. subtilis* (ATCC® 6051™) was selected based on its pathogenic biofilm forming behavior, its generation of bacterial spores as bioindicators of high-stress survival, and its use in prior UV sterilization studies[19-21]. An additional *E. coli* K-12 bacterium, DH5α, was used as a benchmark for planktonic bacterial growth, to simulate pre-seeding growth patterns prior to cell adhesion and biofilm formation.

Using these bioindicators as benchmarks for UVC sterilization effectiveness, we explored three different design concepts to administer sterilizing UV dosages to the internal surface of the ETT. Utilizing ETT cutouts, we tested the effectiveness of UV sterilization to adjust device requirements to achieve an acceptable kill rate and patient safety factor. Our designs include a multi UVC LED stationary illumination probe, a fiberoptic UVC translational illumination probe, and a mono-LED translational illumination probe.

2. METHODOLOGY

2.1 Endotracheal tube coupons

Coupons, with 1 cm diameters, are punched out of sterile endotracheal tubes (Covidien). The coupons are re-sterilized with 70% EtOH for 30 seconds prior to loading onto the pre-sterilized testing base. Coupons are flattened and formed into wells using a washer and two clamps screwed onto the base. A secure coupon acts as the base of a well 1 mm in depth and 7.14 mm in diameter at 1 cm from the objective of the quartz homogenizer. A photograph of sterilized coupons prepared on the base is shown in Figure 1.

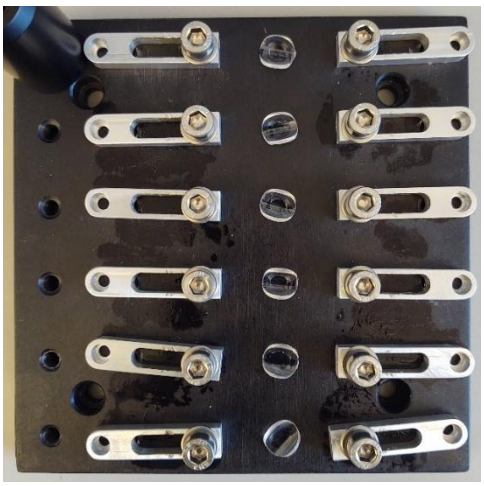

Figure 1. Prepared ETT coupons resting on the testing base prior to well creation by sealing with washers.

2.2 Optical setup

A 50 mW 285 nm UVC LED (THORLABS M285L5, Driver LEDD1B) is held within a custom housing with a hex light pipe to homogenize the light field (Edmund Optics 10x175 mm, fused silica). Light output is measured on a power meter sensor (Newport Model 818-UV) through the well-forming washer determined the power at 1 cm from the objective of the quartz homogenizer to the ETT surface was 1.6 mW. The optical device held above a steel base to attach ETT coupons during testing via screws and stainless-steel washers. All UV sterilization protocols took place in a 37℃ incubator. A photograph of the optical system is shown in Figure 2.

Absorbance tests measured the transmissibility of UVC light (285 nm) by placing a flattened coupon in a washer window over the optical sensor and measuring the transmitted power. The coupon testing windows were made from an ETT pressed between two of the well washers and adhered with tape. A control washer window of the same height and window thickness was used to measure the non-transmitted light. A photograph of an ETT window is shown in figure 3.

Figure 2. Optical setup with coupons secured in wells for UVC testing, located in the incubator.

Figure 3. An ETT coupon pressed between two of the plating washers to measure UVC ETT transmission.

2.3 Inoculum media

Molecular grade water and a biosimilar mucus (3.1% BSA w/v and 5.0% mucin type II w/v in MG H_2O) were used as testing inoculum. Mucus solutions were filter sterilized, made weekly, and stored at 4℃. All inoculums acclimated to room temperature before inoculation. Coupon wells were inoculated with 100 μL of culture inoculum prior to testing. Inoculated coupons underwent a 30-minute acclimation period at 37℃ and either tested immediately or after an additional 3.5 hour settling and binding period.

2.4 Surrogate organisms

Two organisms, *Escherichia coli* (ATCC® 25922™) and *Bacillus subtilis subsp. subtilis* (ATCC® 6051™), were purchased from ATCC as VAP surrogates. An additional culture stock of *E. coli* DH5α was obtained from the University of Washington BIOFAB laboratory for preliminary testing. Stock cultures were incubated on LB plates at 37℃ and 30℃ for *E. coli* and *B. subtilis*, respectively, and stored at 4℃. During testing, single colonies were selected and resuspended in either molecular grade water or biosimilar mucus inoculum. Initial culture concentrations were calculated from 600 nm optical density (OD 600) nanodrop readings and diluted to 0.1 OD prior to testing.

2.5 Ultraviolet illumination

UV sterilization testing took place inside a 37℃ incubator. The objective of the quartz homogenizer was positioned 1 cm above an ETT coupon surface. Dark-colored paper cards were placed to block cross-illumination between coupons during testing. Total illumination time was measured for each coupon. Negative controls of non-UV cultures were used to quantify the UV kill rates. The power of the UVC light was set to 1.6 mW at coupon distance prior to testing each coupon.

2.6 Sterilization effectiveness determination

Surviving bacteria were determined from microbial counts (CFU/mL) following overnight plating. Tested inoculum is drawn up from the well and added to a 1.5 mL tube with 1 mL molecular grade water. The coupon is then picked up with sterile forceps and placed in the 1.5 mL tube and vortexed to resuspend bacteria. 100uL of resuspended tested bacteria are plated on LB plates directly or following a serial dilution to countable colonies. Counts were converted to log values to determine the log reduction (log(CFU/mL)) from control for sterilization effectiveness.

2.7 Multi UVC LED stationary illumination probe design

A design iteration of our photonic device was planned using CAD with physical properties and experimental data. The device features a long and flexible helical polyimide circuit with a string of equally spaced UVC LEDs. The LED circuit is in a flexible UV transmissible silicone housing. The end of the catheter is covered with a non-UV transmissible plastic tip to protect respiratory mucosa. Centering springs allow the catheter to be guided along the central axis of the ETT for equal irradiation of the interior surface. The diameter of the device is 5 mm, smaller than bronchoscopes, and can be used without altering the ETT airflow while the ETT is in use. A generated image of the device can be seen in Figure 4.

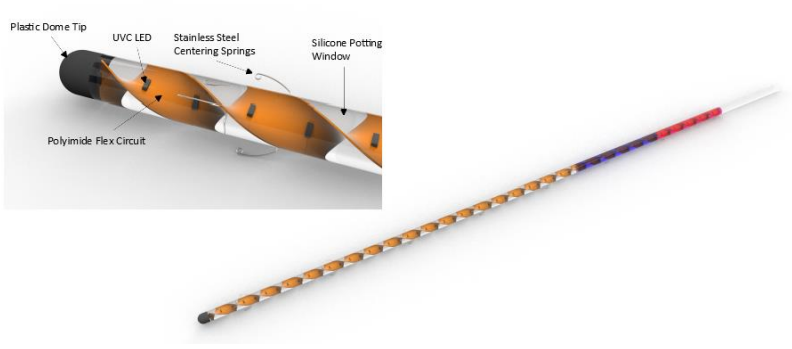

Figure 4. Flexible multi UVC LED stationary illumination probe design is shown with variable illumination lengths of short (orange), medium (+blue), and long (+red) to irradiate different lengths of ETTs.

2.8 Fiberoptic UVC translational illumination probe design

The second design iteration of our photonic device was planned using CAD with physical properties and experimental data. The device operates from an external LED source fed inwards to the ETT via an optical fiber. The fiber is protected with a polyethylene sheath and fed into a brass chassis. The chassis is held by external stainless-steel mounts and fixed to a quartz window. The light can escape the fiber and reflect off an aluminum cone mirror at angles <90°. The mirror and plastic dome tip further protect the vulnerable and delicate respiratory mucosa. A generated image of the device can be seen in Figure 5.

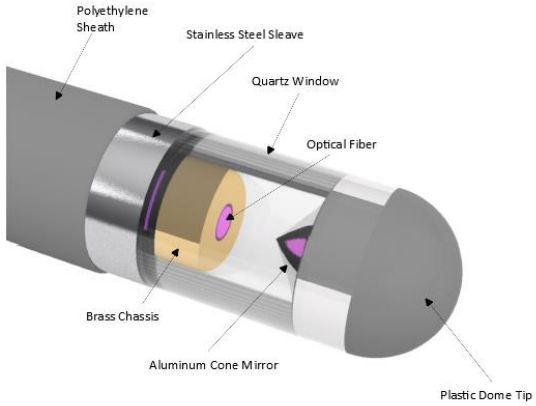

Figure 5. Fiberoptic UVC translational illumination probe powered by external LED source with aluminum reflector and opaque plastic dome tip for safety.

2.9 Mono-LED translational illumination probe design

The third iteration of our photonic UVC catheter was planned using CAD with physical properties and supported by ongoing experimental data. The device operates off a single powerful LED housed within a quartz window on the end of the catheter. This light is reflected off an aluminum cone mirror, distally capped by an opaque dome. The heat generated from the LED is insulated by its ceramic mount while its power source dually provides current and a conductive heat outlet. A generated image of the device can be seen in Figure 6.

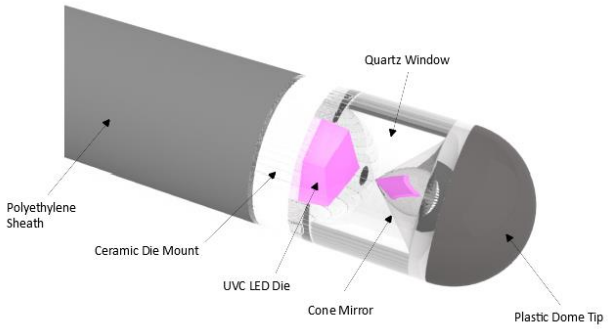

Figure 6. Mono-LED translational illumination probe design with internal LED source with aluminum reflector and opaque plastic dome tip for safety.

3. RESULTS

3.1 ETT UV absorbance

ETT transmittance (UVC LED light centered at 285nm) is measured with different ETT coupon windows with the same window diameter and height as control washer windows. Biosimilar mucus transmission (BMT) is measured with 100 μL of biosimilar mucus deposited on an ETT window. Transmittance in ETTs varied from surface and plastic density

irregularities. Washer control UV indicates antibacterial UV exposure while ETT transmitted UV indicates patient exposure. Percent transmittance indicates the percent of UV light transmitted through the ETT, without biosimilar mucus. UV data is indicated below in Table 3.

The standard daily limit for UV radiation in the 200-315 nm region, given by the American Conference of Governmental Industrial Hygienists (ACGIH), is calculated by the equation: $T_{max}=0.003(J/cm^2)/E_{eff}$, where E_{eff} is the effective irradiance relative to a monochromatic source at 270 nm in W/cm^2 and T_{max} is the maximum exposure time in seconds[22]. Given a safety factor of 1% transmittance, the current level of transmitted UVC light would be considered a safe dosage for 5 minutes.

Table 1. Power measurements of UVC.

Washer control UV	ETT transmitted UV	BMT	Percent transmittance
1.48 ± 0.02 mW	10. ± 5 μW	2.93 ± 0.04 μW	0.7 ± 0.4

3.2 *E. coli* DH5α (planktonic) log reduction – H_2O

0.1 OD 600 *E. coli* DH5α cultures in molecular grade water inoculum, measured with a nanodrop, were placed in ETT coupon wells acclimated to 37°C for 30 minutes in an incubator unit before undergoing illumination with a 285 nm UVC LED. A 4.74-log reduction was achieved after 1 second of UV illumination. A graph of the *E. coli* strain log reductions in variable media can be seen in Figure 7.

3.3 *E. coli* ATCC 25922 (biofilm-forming) log reduction – H_2O

0.1 OD 600 *E. coli* ATCC 25922 cultures in molecular grade water inoculum in ETT coupon wells acclimated to 37°C for 30 minutes in an incubator unit before undergoing illumination with a 285 nm UVC LED. A 1.72-log reduction was achieved after 1 second of UV illumination. A graph of the *E. coli* strain log reductions in variable media can be seen in Figure 7.

3.4 *B. subtilis* ATCC 6051 (biofilm-forming) log reduction – H_2O

0.1 OD 600 *B. subtilis* ATCC 6051 cultures in molecular grade water inoculum in ETT coupon wells acclimated to 37°C for 30 minutes in an incubator unit before undergoing illumination with a 285 nm UVC LED. A 0.81-log reduction was achieved after 1 second of UV illumination. A graph of the *B. subtilis* log reduction in H_2O can be seen in Figure 7.

3.5 *E. coli* ATCC 25922 (biofilm-forming) log reduction – Biosimilar mucus

0.1 OD 600 *E. coli* ATCC 25922 cultures in biosimilar mucus inoculum in ETT coupon wells acclimated to 37°C for 30 minutes in an incubator unit before undergoing illumination with a 285 nm UVC LED. No log reduction was achieved after 1 second of UV illumination. A graph of the *E. coli* strain log reductions in variable media can be seen in Figure 7.

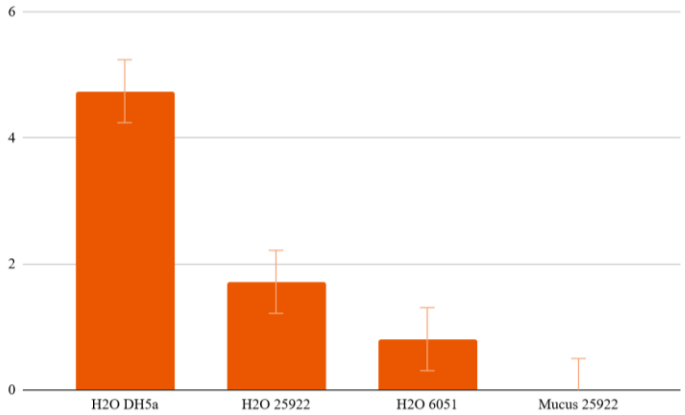

Figure 7. Log reduction of bacterial species to media type with 1 sec. UV exposure following a 30-minute 37°C acclimation.

3.6 *E. coli* ATCC 25922 (biofilm-forming) log reduction – Biosimilar mucus, variable time

0.1 OD 600 *E. coli* ATCC 25922 cultures in biosimilar mucus inoculum in ETT coupon wells acclimated to 37℃ for 4 hours in an incubator unit before undergoing illumination with a 285 nm UVC LED. Duration of UV varied between data sets, with times of 1, 10, 30, 60, and 120 seconds of illumination to control. No significant log reduction was achieved after any duration of UV illumination, as shown in Figure 8.

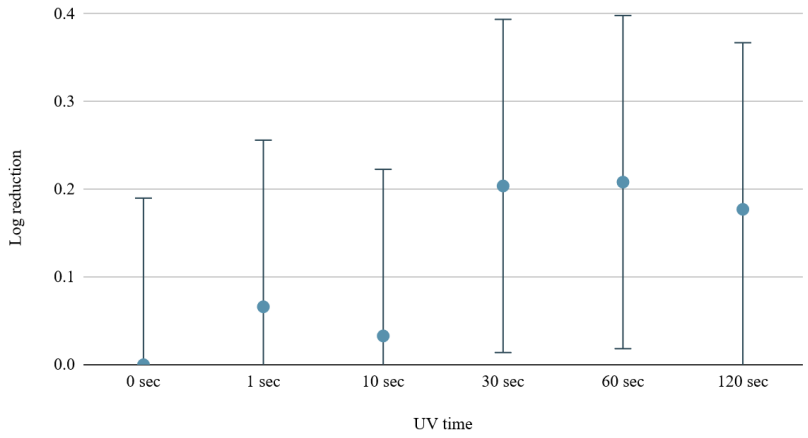

Figure 8. Log reduction of *E. coli* 25922 in biosimilar mucus inoculum following a 4-hour 37℃ incubation on an ETT surface and shown with variable UV duration.

4. DISCUSSION

Use of 285 nm ultraviolet light proved to be ineffectual to sufficiently reduce VAP surrogate organisms at high log reduction values. Much of the data gathered shows insignificant log reduction due to one or many factors, such as non-diffuse UVC illumination, bacterial clumping, and meniscus effects in ETT coupon wells. While the planktonic, lab-acclimated *E. coli* DH5α did show potentially significant log reduction, the VAP surrogates did not behave similarly. This is especially notable in cultures tested in biosimilar mucus inoculum, where there was no significant log reduction. BSA, a component of the biosimilar mucus, has been shown to promote cell adhesion by promoting bacterial/surface interactions through mediating binding[23]. Cell to surface adhesion is an important step in the formation of a biofilm. Biofilms have been found to have remarkable UV and antibacterial agent resistance[24]. While the pre-testing incubation time of 30 minutes is not long enough to form biofilms, the lengthier 4-hour incubation time could allow for the settling and acclimation of bacteria to the environment. Large proteins, like BSA, and DNA absorb UV light within the UVC range[25,26]. While this is the reason for UVC's germicidal effects, the biosimilar mucus also acts as a shield for radiation on the pathogenic bacteria. Shorter wavelength UVC illumination, longer exposure time, and/or higher UV power would be needed to pass through the mucus barriers and kill the bacteria[27].

Our photonic catheter schematics explore the design space between the safety factors and UVC illumination requirements for successful log reductions. Most importantly, we consider the safety of the patient to be of the highest concern, as UVC light can lead to acute tissue damage or chronic conditions like cancer. The current levels of UVC transmittance through the ETT fall well below the safety guidelines of epidermal and corneal exposure during the UV sterilization protocol[22]. Ease of use for hospital staff was an important factor in designing the catheter units since VAP bundle hygienic protocols are heterogeneously adopted[8]. Integrating these two factors, we will continue to explore UVC LED power and illumination time ratios to optimize patient safety, ease of use, and biofilm-prevention.

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