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Application of the novel environment-cleaned light-emitting diode devices in various test fields

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Abstract

Faced with the impact of extreme climate, countries have proposed carbon dioxide emission reductions, hoping to achieve carbon neutrality by 2050, and Taiwan's industries must also face the transformation of low-carbon green energy. Light-emitting diode (LED) is a cost effective semiconductor device that produces light within a narrow bandwidth of wavelength through electroluminescence. Recently, LED technology has attention to apply in the area of food production, preservation, and safety. At present, some researches have been demonstrated that the antimicrobial LED visible light is less anti-microbial efficacy than ultraviolet (UV) light. However, the antimicrobial LED visible light has been recognized as an alternative technology to UV light since it is an environmentally friendly and safe technology for human and animals. For this reason, LED technology has recently received attention for applying in many test fields as laboratory, pig farms, computer, bio-medical industries etc. In this study, this novel clean and disinfect tool-novel environment-cleaned LED devices were tested in the various fields and obtained the positive results as the application of novel environment-cleaned LED devices on anti-fungal efficacy, and ethylene, $PM_{2.5}$, and harmful gas degradations in laboratory, anti-bacterial and virus efficacy in laboratory and/or pig farms, anti-microbial notebook panel development, and anti-colorectal cancer *in vitro*. In the future, we wish this novel environment-cleaned LED devices will friendly used in human and animal environments to decrease the harmful matters in the environments.

Keywords: Antimicrobial Efficacy; Atmospheric Fine Particulate Matter; Degradation; Harmful Gas; Fields; Light-Emitting Diode

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1. Introduction

Ultraviolet (UV) rays are divided into UV-A (320-400 nm), UV-B (280-320 nm), and UV-C (100-280 nm) according to the wavelengths. Among them, UV-C light outside the wavelength can destroy the DNA or RNA molecular structure of microbial cells by emitting UV light, so that bacteria or viruses can die and achieve the effects of sterilization and disinfection. According to the studies, UV-C light with a wavelength of about 250-260 nm has the strongest bactericidal effect. UV light is a powerful light for surface decontamination. However, UV light has many limitations at harmful effects for human and animals [1-6]. Previously, the germicidal efficacy of 405 nm wavelength light is significantly lower than that of UV light [1-6]. The anti-microbial light at 405 nm wavelength can be used for the surface decontamination to avoid affecting the exposed mammalian cells [1-6]. At present, the UV light source used for medical sterilization mainly comes from UV mercury lamp. The mercury lamp adds mercury into the vacuum quartz lamp tube, discharges through the voltage difference between the two ends of the electric shock, excites the UV radiation wave, and then changes the wavelength through the fluorescent paint inside the lamp tube.

The sterilization effect of UV light depends on the radiation intensity (and illuminance) of the UV light and the irradiation time, which are combined into the irradiation dose. The stronger the illuminance is related with the shorter the required irradiation time for the sterilization. On the contrary, if the illuminance is low, a longer irradiation time is required to meet the same light dose and achieve the effect of sterilization and disinfection [7-9]. At present, the main reason why UV-C LEDs cannot completely replace the sterilizing UV mercury lamps for medical use is that the illumination of LEDs is still not strong enough. To achieve the same sterilization effect as mercury lamps, a larger number of LEDs or more irradiation time will be required. For the professional medical units with high frequency of use and a large range of sterilization spaces, it is not cost-effective to use UV-C LEDs for disinfection at this stage. However, for the general daily sterilization, UV-C LEDs have gradually begun to help. Compared with the UV mercury lamp, the light source of UV-C LEDs is smaller in size and can be used in different applications. At present, the most common applications are mainly air sterilization, static sterilization, and flowing water sterilization [10-15].

Cancer involved progresses through multiple stages as initiation, promotion, progression, and metastasis [16]. Currently, colon cancer is very important cancer worldwide. Although many therapies and therapeutic strategies are applied in the cancer patients, however, the deaths of patients with cancer maintain in the top of the dead people worldwide [17]. Therefore, novel therapies and therapeutic strategies are needed for the cancer therapy in the future. Previously, the photodynamic therapy has been increasingly used in clinical medicine via the visible light at a wavelength corresponding to the drug absorption activates these agents and induces highly cytotoxic products [18-20].

The ethylene is well known for its role in promoting the ripening of bananas, tomatoes and other fruits. Plants can produce ethylene and use it to regulate growth and development processes such as seed germination, senescence, and shedding of flowers and leaves, and programmed cell death. After picking, the fruit will be overripe due to the production of a large amount of ethylene, which will greatly shorten the storage period and shelf life. The adverse weather factors and serious pests and diseases will induce crops to produce a large amount of ethylene and lead to premature decline in yield, which will bring a great deal to the loss of agricultural production. Therefore, how to regulate amount of ethylene in fruit is an important issue [21].

The computer is an important tool in modern and is also the daily routine of all office workers. When they come home from get off work, they switch to a mobile phone and keep sliding. Because of the needs of work and life, modern people often use 3C devices included mobile phones, tablets, and desktop computers. Therefore, modern people use their eyes all the time, which can easily lead to various eye diseases as inflammation, red eyes, discomfort etc. Finally, these various eye diseases will affect work. Therefore, the computer has anti-blue light function that can reduce the damage to our eyes included cornea, lens, and retina. In addition, the design of its backlight source which can reduce the chance of screen splash. If the screen flickers, it will cause eye fatigue. If the screen does not flicker, it can provide the comfort of our eyes. On the other function, the screen has an anti-microbial function which can reduce our touch and the infection caused, especially the children like to touch everywhere, so that parents can feel more at ease. Therefore, the research and development of anti-microbial screens for the modern people to use for a long time is very important [21].

In the traditional swine and poultry farms, the disinfectants are used for the cleaning and disinfection regimen. Disinfectants are used on swine and poultry farms for disinfecting the surfaces of floors, walls, equipment, and tools. The ideal detergents should be left no residue and it should be non-toxic to pigs, poultry, and operative staffs and it must have minimal environmental impacts. Therefore, in order to avoid detergent residues and possible effects on pigs and poultry, operative staffs, and environment, the research and development of a novel method for anti-microorganisms and other harmful substances in the pig and poultry farms is important.

2. Material and methods

2.1 Cells and Culture Condition

HT-29 (ATCC® HTB-38™) was purchased from ATCC (Manassas, VA 20110). McCoy's 5a medium, fetal bovine serum (FBS), and antibiotics (penicillin and streptomycin) were purchased from Sigma-Aldrich. HT-29 cells were cultured in McCoy's 5a medium. McCoy's 5a medium was supplemented with 10% FBS and 1% penicillin and streptomycin. The cells were incubated at 37°C with 5% CO_2. Cells were sub-cultured to replace flesh media per 2-3 days when they became confluent.

A monkey kidney cell line used was MARC-145 (ATCC® CRL-12231™). MARC-145 cells were grown in Dulbecco's modified Eagle's medium (DMEM; GIBCO®) supplemented with 10% fetal bovine serum (FBS; HyClone®), 2 mM L-glutamine (Invitrogen®), 100 U/mL penicillin and 100 mg/mL streptomycin (Invitrogen®) in a humidified 5% CO_2 incubator at 37°C.

2.2 Bacterial Strains

Eight *Salmonella* spp. strains (ATCC® 10743™, ATCC® 10744™, ATCC® 10746™, ATCC® 10747™, ATCC® 13852™, ATCC® 14878™, ATCC® 14879™, and ATCC® 17495™) were used in this study. These bacteria were grew on XLD agar under the 37°C and aerobic status.

2.3 Source of Apples and Virus

Apples were order from the supermarket, Miaoli, Taiwan. Porcine reproductive and respiratory syndrome virus (PRRSV) (strain MD-005) was kindly shared from Dr. Wen-Bin Chung (National Pingtung University of Science and Technology).

2.4 Experimental Grouping

In this study, two group (the normal LED group and the functional group) were divided in each experiment. All experiments were performed in Division of Animal Industry, Animal Technology Research Center, Agricultural Technology Research Institute, Taiwan.

2.5 Viral Challenge Test

The Taiwan local strong virulence of PRRSV (strain MD-005, viral titer is 10^5 $TCID_{50}$/mL) was challenged to MARC-145 cells. At the each designed experimental points, the samples were collected to detect the cycle threshold value (Ct value).

2.6 Cell Viability Assay

HT-29 cells (5×10^4/mL) were initially incubated for 24 h in a 96 well plate, respectively. The lighting time in HT-29 of each group (the normal LED group and the functional LED) were 0, 0.5, 1, 2, and 4 hour, respectively. At each time point, cell viability was detected by MTT cell viability assay kit [3-(4, 5-Dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide; MTT], was purchased from abcam®. The reduced purple dye intensity of color was estimated by reading at optical density 570 nm in a spectrophotometer.

2.7 Microbial Incubation

Tryptone Soy Agar (TSA) was used to perform air environmental microbiological monitoring. TSA plates were placed in the 37°C after collecting the microbial samples. After 18-24 hours incubation, the number of colony-forming units (CFU) and the percentage of microbial growth rate (%) were calculated.

2.8 Monitor of Air $PM_{2.5}$, NH_3, H_2S, Air Microbial Concentrations, and Pigs' Clinical Behaviors after LED lighting in Pig farm

In this study, the pigs' clinical behaviors were monitored by a senior veterinarian. Six indexes of clinical behavior as spirit, appetite, excretion, breathe, gait, and body appearance are used for the score (Table 1). The detection of air $PM_{2.5}$, NH_3 and H_2S, and air microbial concentrations were performed by using Multifunction Air Detector (WP6912; VSON®, Shenzhen, China) and AP Buck BioCulture Microbial Air Sampler (discontinued) (buck Bio-Culture-B30120; A.P. BUCK INC., Orlando, Florida, USA), respectively.

Table 1 Six indexes of clinical behavior as spirit, appetite, excretion, breathe, gait, and body appearance for the score

Score	Spirit	Appetite	Excretion	Breathe	Gait	Body appearance
1	Normal	Normal	Normal	Normal	Normal	Normal
2	Inactive / weak	Suboptimal	Atherosclerosis	Slight	Slight limp	Petechial bleeding / Scabs
3	Lying down	Unable to eat	Watery diarrhea	Severe	Severe limp	Anemia / Jaundice

2.9 Detection of Fungal Area on the Apple Sections and the Degradation of Ethylene Concentration in Laboratory

In this study, the fungal area on the apples and the degradation of ethylene concentration were detected in Laboratory. Ethylene concentration was detected by using MacView Ethylene Postharvest Portable Analyser (HACPHA000102233; MacView®, Smartec Scientific Corp., Taipei, Taiwan).

2.10 Monitor of Bacterial Concentrations on the Computer Screen

In this study, the detection of bacterial concentrations on the computer screen was performed via researcher's fingers and microbial incubation. After 18-24 hours incubation, CFU and microbial growth rate (%) were calculated.

2.11 Statistical Analysis

The data were expressed as mean ± SD. All comparisons were made by one-way ANOVA and all significant differences are reported at $^{*}p < 0.05$, $^{**}p < 0.01$, and $^{***}p < 0.001$.

3. Results

3.1 Application of LED Devices on Anti-Fungal Efficacy and Ethylene Degradation

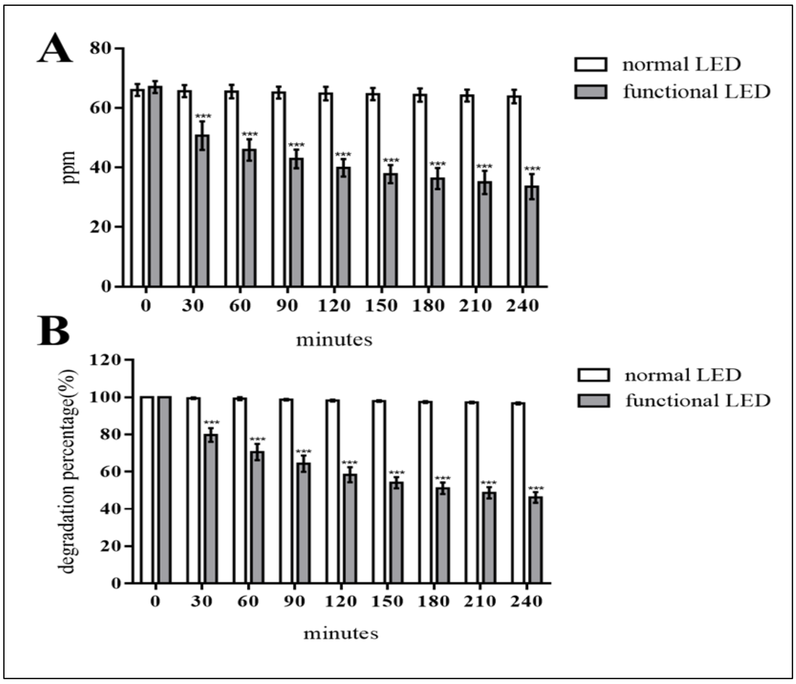

Figure 1 Application of functional LED devices (450 nm wavelength) on ethylene degradation. (A) The concentrations of ethylene (ppm). (B) The degradation percentages (%) of ethylene. The values are mean ± SD. $^{***}p < 0.001$

Two groups (the normal LED group and the functional LED group) were divided in this experiment in order to verify the efficacies of functional LED on anti-fungus and ethylene degradation. The functional LED devices at 450 nm wavelength significantly decreased ethylene concentrations in a time-dependent manner. After the functional LED lighting for 30-240 minutes, the ethylene concentrations significantly decreased ($p < 0.001$) (Figure 1A). The degradation percentages (%) of ethylene were also significant decrease ($p < 0.001$) between 30-240 minutes LED lighting. After the functional LED lighting for 30 and 240 minutes, the degradation percentages (%) of ethylene were about 20% and over 40%, respectively (Figure 1B).

In addition, the efficacy of functional LED on anti-fungus was evaluated via apple section-lighting assay. Based on the results, the functional LED devices at 450 nm wavelength significantly decreased fungal area on the apple section in a time-dependent manner. After the LED lighting for 4-7 days, the fungal area (mm^2) on the apple section under the functional LED lighting more decreased than the normal LED lighting (Figure 2).

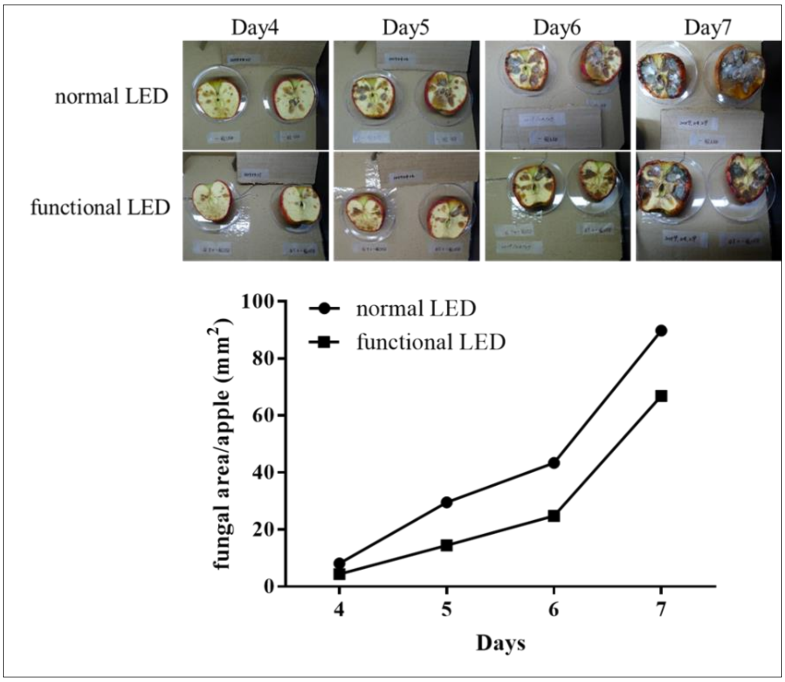

Figure 2 The efficacy of functional LED on anti-fungus was evaluated via apple section-lighting assay. After the LED lighting for 4-7 days, the fungal area (mm^2) on the apple section under the functional LED lighting more decreased than the normal LED lighting

3.2 Application of the Functional LED Devices on Anti-Bacterial Efficacy

The efficacy of functional LED devices on anti-bacteria was evaluated via detecting the bacterial concentrations in air and inhibiting the bacterial growth on agar. Based on the results, the functional LED devices at 450 nm wavelength significantly decreased bacterial concentrations in air ($p < 0.05$). Moreover, the bacterial growth of eight *Salmonella* spp. strains (ATCC® 10743™, ATCC® 10744™, ATCC® 10746™, ATCC® 10747™, ATCC® 13852™, ATCC® 14878™, ATCC® 14879™, and ATCC® 17495™) was more suppressed under the functional LED lighting than the normal LED lighting (Figure 3).

3.3 Application of the Functional LED Devices on Anti-Virus Efficacy

The efficacy of functional LED on PRRSV was evaluated via detecting Ct values. Based on the results, the functional LED devices at 450 nm wavelength significantly decreased Ct values of PRRSV ($p < 0.001$) compared to the normal LED devices (Figure 4).

3.4 Evaluation of the LED beads of the Functional LED Devices for Anti-Bacterial Efficacy

The LED bead efficacy of functional LED devices on anti-bacteria was evaluated via inhibiting the bacterial growth on agar. Based on the results, the half LED beads, one third LED beads, and 100% LED beads of the functional LED devices at 450 nm wavelength all significantly decreased the bacterial concentrations in air for 16-24 hours lighting in laboratory study (Figure 5).

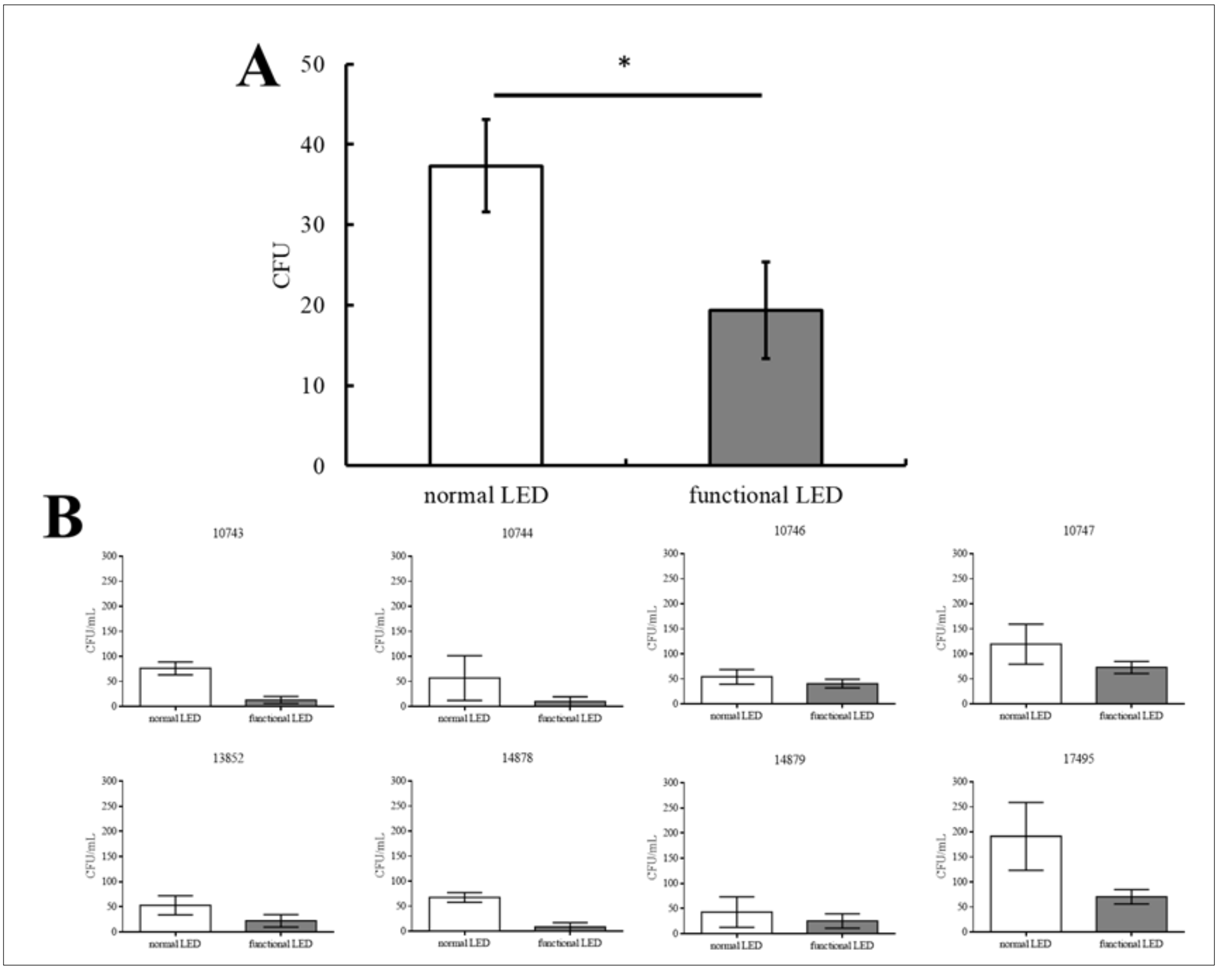

Figure 3 Anti-bacterial efficacy via the functional LED devices. (A) Bacterial concentrations in air. (B) Eight *Salmonella* spp. strains (ATCC® 10743™, ATCC® 10744™, ATCC® 10746™, ATCC® 10747™, ATCC® 13852™, ATCC® 14878™, ATCC® 14879™, and ATCC® 17495™). The values are mean ± SD. $^{*}p < 0.05$.

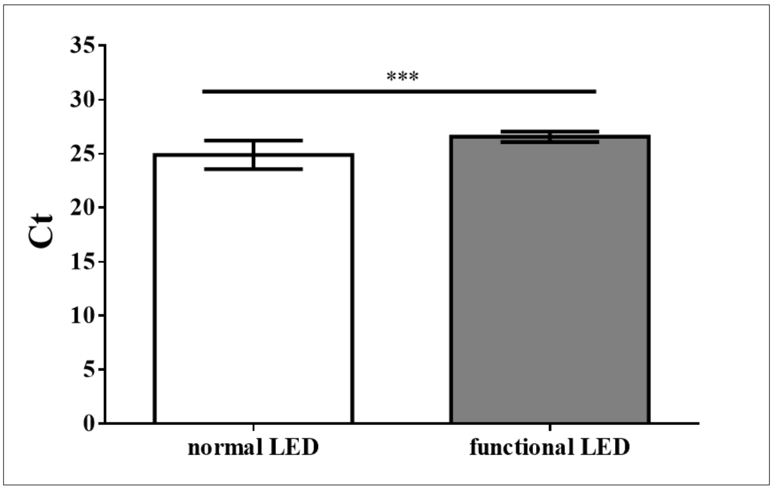

Figure 4 Anti-porcine reproductive and respiratory syndrome virus efficacy via the functional LED devices. The Ct values are mean ± SD. $^{***}p < 0.001$

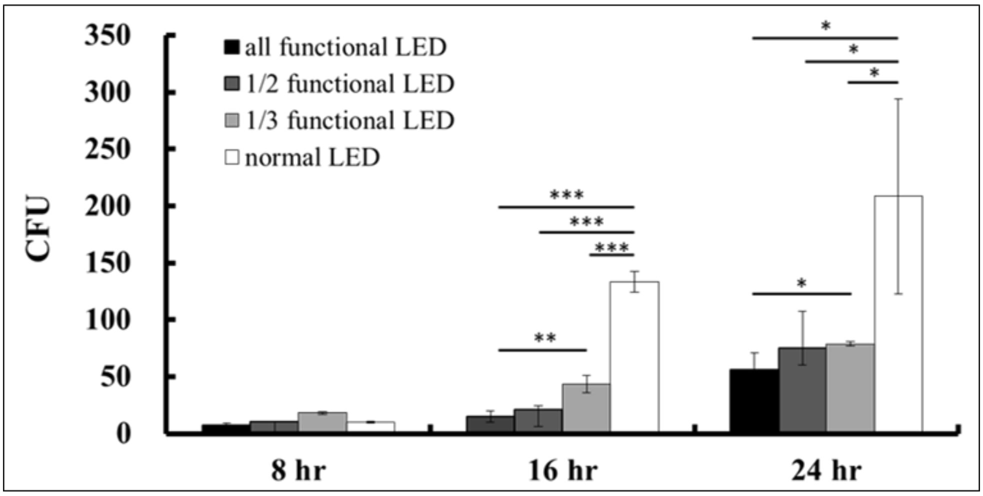

Figure 5 Anti-bacterial efficacy via the functional LED devices with different LED bead numbers. The values are mean ± SD. $^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$

3.5 Application of the Functional LED Devices for $PM_{2.5}$ Degradation

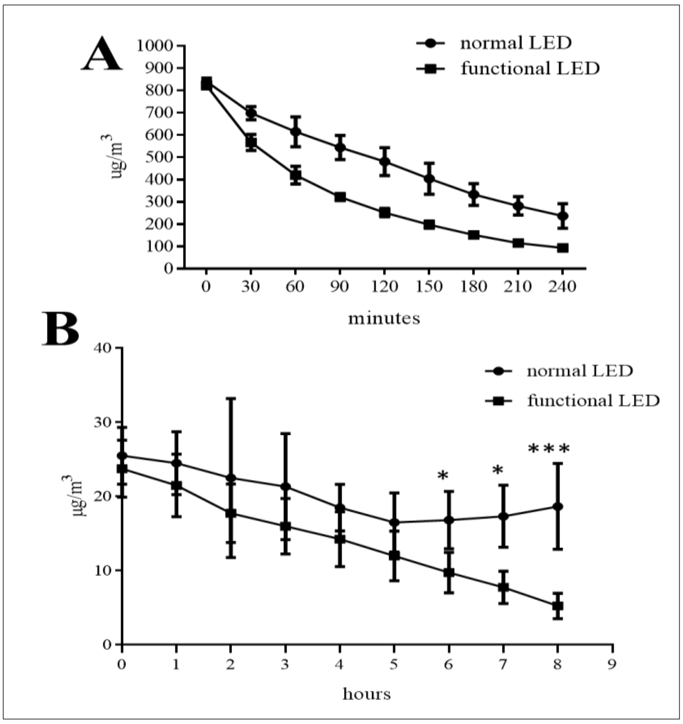

Figure 6 Application of the functional LED devices for $PM_{2.5}$ degradation. (A) $PM_{2.5}$ degradation via the functional LED devices in laboratory study. (B) $PM_{2.5}$ degradation via the functional LED devices in pig field trail. The values are mean ± SD. $^{*}p < 0.05$; $^{***}p < 0.001$

The degradation concentration of $PM_{2.5}$ was evaluated in laboratory study and pig field trail by the functional LED lighting. Based on the results, the functional LED devices at 450 nm wavelength significantly decreased $PM_{2.5}$ concentrations in laboratory study in a time-dependent manner (Figure 6A). Moreover, the same effects were presented in the pig field trail. After 6 hours LED lighting in the pig field trail, the functional LED devices significantly decreased the concentration of $PM_{2.5}$ in air in the pig farm (Figure 6B).

3.6 Application of the Functional LED Devices for Harmful Gas Degradations

The degradation concentrations of NH_3 and H_2S were evaluated in pig field trail by the functional LED lighting (Figure 7). Based on the results, the functional LED devices at 450 nm wavelength significantly decreased NH_3 and H_2S

concentrations in pig field trail in a time-dependent manner (Figure 8). After 4-5 hours LED lighting in the pig field trail, the functional LED devices significantly decreased the concentration of NH_3 and H_2S in air in the pig farm (Figure 8).

Figure 7 Application of the functional LED devices in a traditional pig farm. (A-B) The field trail. (C-D) LED lighting test

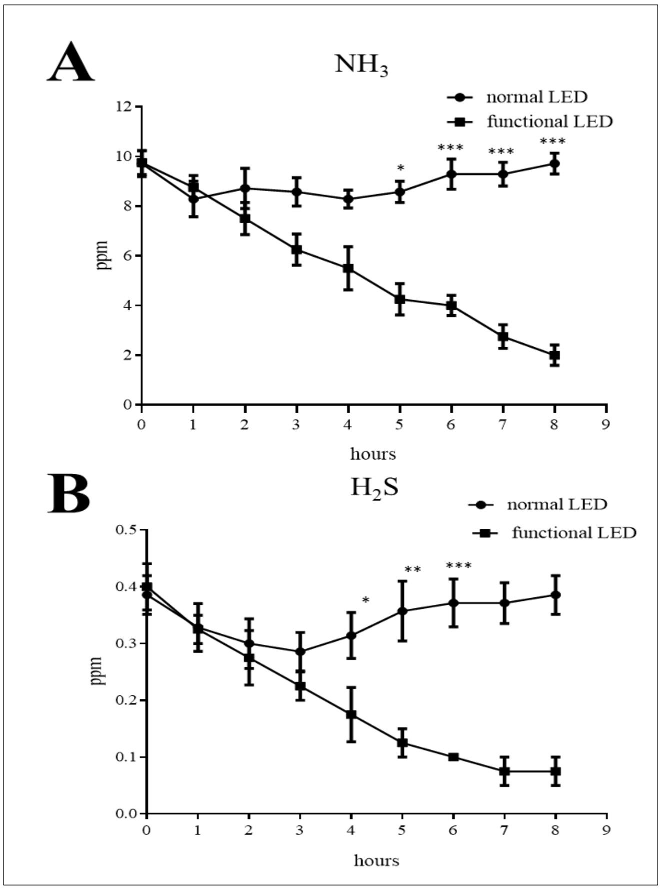

Figure 8 Application of the functional LED devices for harmful gas degradations. (A) NH_3. (B) H_2S. The values are mean ± SD. $^{*}p < 0.05$; $^{**}p < 0.01$; $^{***}p < 0.001$.

3.7 Application of the Functional LED Devices for Anti-Microbial Computer Screen Development

The efficacy of functional LED devices on anti-bacteria was evaluated via detecting the bacterial concentrations on the computer screen. Based on the results, the functional LED devices at 450 nm wavelength significantly decreased bacterial concentrations on the computer screen ($p < 0.01$) (Figure 9A). Moreover, during the 1 minute to 4 minutes-finger touch computer screen, the bacterial growth of finger was more suppressed under the functional LED lighting than the normal LED lighting (Figure 9B).

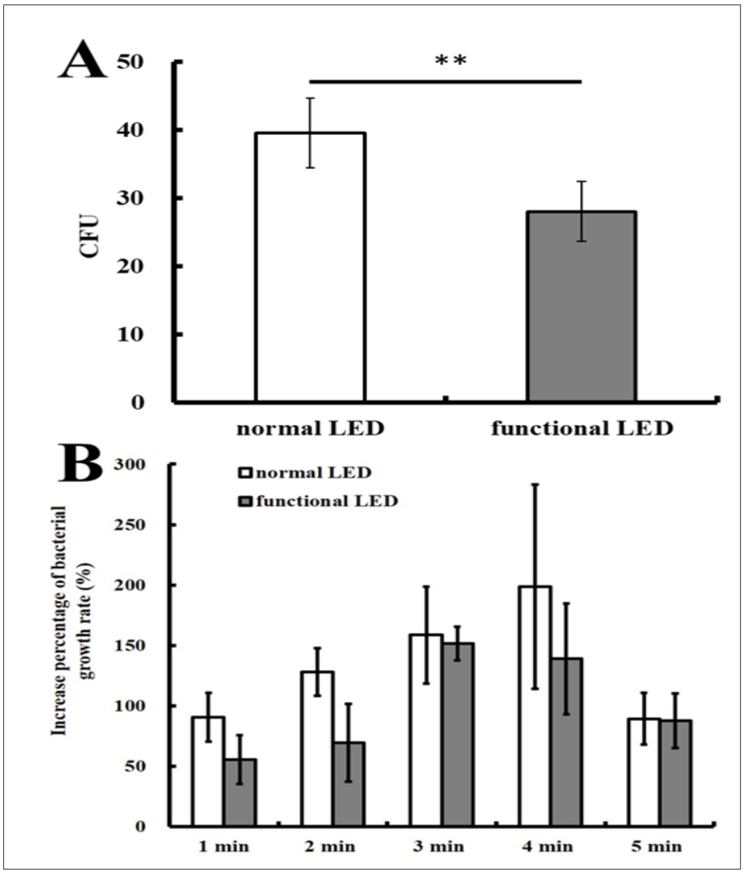

Figure 9 Application of the functional LED devices for harmful gas degradations. (A) NH_3. (B) H_2S. The values are mean ± SD. $^{**}p < 0.01$

3.8 Application of the Functional LED Devices for Anti-Colorectal Cancer *in vitro*

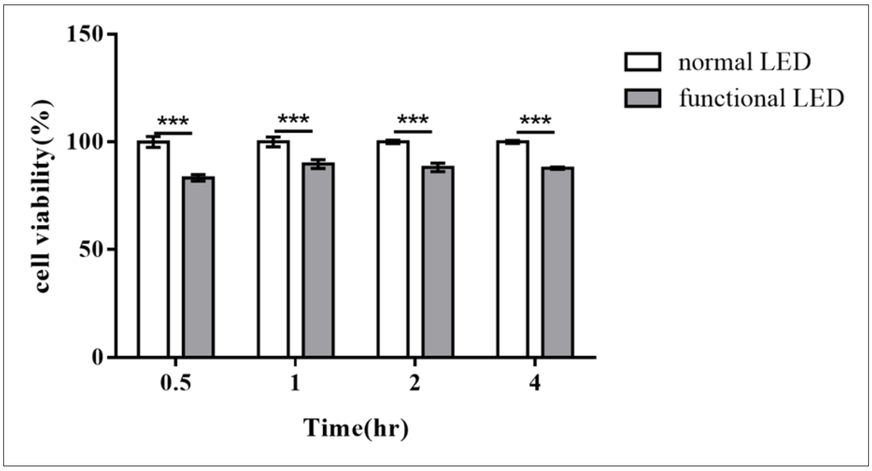

Figure 10 Application of the functional LED devices for anti-colorectal cancer *in vitro*. The values are mean ± SD. $^{***}p < 0.001$

The efficacy of functional LED devices on anti-colorectal cancer *in vitro* was evaluated. Based on the results, the functional LED devices at 450 nm wavelength significantly inhibited the growth of colorectal cancer cell line HT-29 ($p < 0.001$) during 0.5-4 hours lighting (Figure 10).

4. Discussion

The technology of LED is becoming more and more mature and there are more and more fields of application. Some applications of LED in lighting includes residential areas as wall lamps, night lamps, auxiliary lighting, garden lamps, and reading lamps etc.; facilities as emergency indicator lights, and hospital bed lights etc. ; store areas as spotlights, lanterns, barrel lights, light strips etc.; outdoor areas as building exteriors, solar lights, and light show etc. [21].

The lighting market has always been regarded as the largest and most promising market for LEDs, although currently due to cost and product characteristics constraints, major lighting products cannot be launched. But it cannot be denied that due to the rapid development of LED technology in recent years, some market shares have been achieved in the special lighting market. In the general lighting market, products include barrel lamps, socket lamps, projection lamps, and landscape lighting have also been launched one after another, and some products have begun to replace traditional light sources such as halogen lamps and incandescent bulbs, allowing highly optimistic attitude of manufacturers to embrace the future development of LED lighting [22-24].

In the context of the rapid improvement of the LED function/price ratio, as well as the continuous increase in energy prices and environmental protection pressure, the LED lighting market will grow rapidly. The key to the growth of the market scale lies in the popularization of the application of high-brightness and high-power LEDs, the improvement of lifespan, the improvement of color rendering, the improvement of luminous efficiency, the reduction of prices, and the formulation of LED lighting specifications. LED is facing the crossroads of new applications. How to effectively develop new applications and build consumer confidence in the quality of LED lighting products is the most important key to opening up new markets [21, 24].

Porcine reproductive and respiratory syndrome (PRRS) is a very important swine disease and this disease is difficult to clear via many methods. PRRSV infection in pigs mainly causes miscarriage and stillbirth of sows and respiratory tract lesions of nursery pigs, which seriously affects the economic loss of pig farmers. Microbial inactivation via LED technology is commonly referred to as photodynamic inactivation by producing reactive oxygen species (ROS) to kill pathogens. According to our recent research, the novel LED devices possess $PM_{2.5}$ and harmful gas degradation, and antimicrobial efficacy. This novel clean and disinfect tool will be applied in the pig farms in the future to decrease disease occur and reduced economic losses in pig farms [25-41].

Photodynamic inactivation (PDI) via the visible light inactivation is a non-thermal photophysical and photochemical reaction that requires visible light [42-43]. Application of PDI for preventing pathogens was in many fields as food production, food preservation, food safety, and fruit storage etc. [42-43]. Additionally, scientists have identified some key regulatory components in the process of ethylene signal transduction for more than 20 years research. Among of key regulatory components, ethylene-insensitive protein 2 (EIN2) is the core positive regulator of plant response to ethylene, and its functional loss will lead to complete loss of ethylene response in plants. According to our results, our novel LED devices possess antimicrobial efficacy and ethylene degradation efficacy. Therefore, our novel LED devices can be severed as a sharp weapon for food preservation [42-43].

The spread of the COVID-19 epidemic h1as affected the global economy. Countries have adopted different anti-blocking strategies and introduced effective defense mechanisms. Using COVID-19 to warn the world of the importance of health protection concepts and environmental protection awareness, it has also prompted enterprises to think deeply and accelerate necessary transformation. A crisis is also a turning point. How to strengthen the safety protection level of the owner's wealth-producing equipment and check for consumers? How to improve the overall operation efficiency of the enterprise and protect the health of employees? Epidemic prevention measures can not only be considered for a while, but can also improve operation efficiency, consolidate the foundation of the enterprise, and long-term operation. It is the most important task for enterprises to transform under the epidemic situation. During the epidemic, reducing non-essential contact can help reduce the chance of infection. In order to reduce the chance of contact, self-service equipment has become the first choice of consumers for shopping and dining. Self-service provides a safe distance maintenance and improves consumer autonomy. However, the antibacterial cleaning of touch screens is also important and can prevent pollution and odor-causing bacteria.

5. Conclusion

Many functional features have demonstrated in our novel LED devices includes degradation of atmospheric fine particulate matter ($PM_{2.5}$), harmful gases (NH_3 and H_2S), and antimicrobial efficacy. Currently, this novel LED devices have already promoted to several application fields. In the future, we hope this novel LED devices have the promising applications in the human and animal life environment and biomedical researches.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors declare no conflict of interest.

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