Research Article Vol. 88, No. 10 / October 2021 / Journal of Optical Technology 587

Journal of Optical Technology

Shortwave excilamps as effective sources of radiation for inactivation of viruses and bacteria

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Received 29 June 2021; Opticheskiĭ Zhurnal **88**, 50–58 (October 2021)

The modern level of research on the inactivation of viruses, bacteria, and living cells under the influence of ultraviolet radiation from excilamps is presented. Special attention is paid to the inactivation of viruses, which is relevant in connection with the spread of coronavirus infections. It has been shown that UVC excilamps, emitting in the wavelength range of 200–240 nm, are an alternative to classical sources of ultraviolet radiation for inactivating viruses. They can be used alone or in combination with known sources (e.g., LEDs, low pressure mercury lamps). A forecast was made for the development of a new technology for disinfecting air and surfaces from infectious agents of various etiologies.

OCIS codes: (000.1430) Biology and medicine, (230.6080) Sources, (260.7190) Ultraviolet, (350.5130) Photochemistry.

https://doi.org/10.1364/JOT.88.000587

1. INTRODUCTION

Excilamps are a generic name for a class of optical devices that emit spontaneous ultraviolet (UV) and/or vacuum ultraviolet (VUV) radiation from excimer and exciplex molecules. Traditionally, they are classified both by types of working molecules and by the method of excitation of the gaseous medium and design [1–4]. Dielectric-barrier discharge (DBD) excilamps are the most widely used today, and the first patent for a DBD excilamp was obtained at the S. I. Vavilov State Optical Institute in St. Petersburg, Russia [5]. Over the past 25 years, the Institute of High Current Electronics of the Siberian Branch of the Russian Academy of Sciences (IHCE SB RAS) has accumulated unique expertise in the study of excilamps and the creation of irradiation installations [3,4,6]. As shown below, part of this expertise came to be critical in solving the problem of protecting the population from known and new lethal respiratory infections.

The purpose of this work is to give a systematic understanding of the scientific foundations and applications of excilamps in the creation of irradiation installations for the disinfection of air and surfaces from viral and bacterial particles. The relevance of research is due to both the increasing resistance of common pathogens to antibiotics and an increase in the viral background due to the emergence of new, lethal respiratory infections, such as severe acute respiratory syndrome (SARS, 2003), Middle East respiratory syndrome (MERS, 2012), and COVID-19 (2019–2020), caused by the corresponding coronaviruses SARS-CoV, MERS-CoV, and SARS-CoV2. This demands the development of new devices for increased efficiency of air disinfection in rooms and on surfaces, as well as in gas and liquid media.

2. CLASSICAL APPROACH TO UV INACTIVATION OF MICROORGANISMS

It is known that UV radiation with wavelengths $\lambda = 205-295$ nm has an inactivating effect (i.e., it slows down vital processes until their complete suppression) on bacterial cells [7,8], which is widely used to create so-called irradiating germicidal installations. This radiation is absorbed by both purine (adenine, guanine) and pyrimidine (cytosine, thymine, uracil) nitrogenous bases of DNA. The latter, as proved by Gates [7], is the main target for lethal and mutagenic effects of UV radiation. For example, Fig. 1 shows the spectrum of the inactivating effect of UV radiation on the bacterial culture of *Escherichia coli* [9] (curve *2*).

Note that, within the bactericidal range of the spectrum, sensitivity to the action of UV radiation of viruses and cells of various origins can vary greatly. Irradiation always produces various mutant types: some can be considered completely inactivated because their ability to replicate has disappeared, and in mutants of other types, inactivation of some cellular subsystems takes place, but not the system altogether. Moreover, if the device that restores the disturbances remains intact, after a while, the system can become viable again and even acquire stability (resistance) to radiation.

To obtain high-quality disinfection, it is also important to consider that some subsystems of cells have the ability of photoreactivation [10,11]. For example, the doses required to ensure 99.9% inactivation of various microorganisms that have also been subjected to photoreactivation by ultraviolet radiation of the A range (UVA) ($315 < \lambda < 400$ nm) can be 2–4 times

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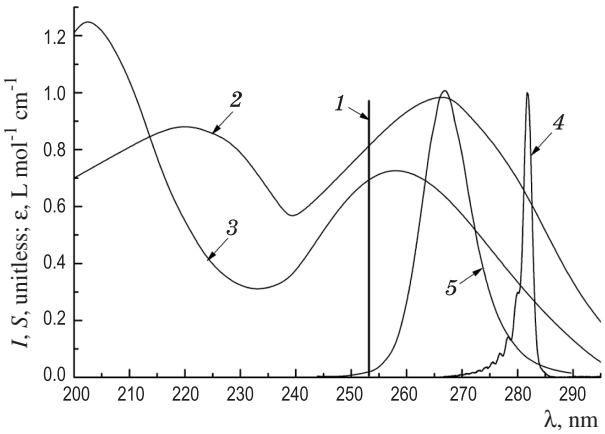

Fig. 1. Spectral characteristics of UVC radiation sources and biological objects. *1*—atomic line of mercury in a low pressure mercury lamp, *2*—action spectrum for inactivation of UV radiation on *Escherichia coli*, $S(\lambda)$ [9], *3*—absorption spectrum of DNA, $\varepsilon(\lambda)$ [13], *4*—emission spectrum of the $B \rightarrow X$ band of the XeBr barrier discharge excilamp, *5*—radiation spectrum of a UVC LED [14].

higher than microorganisms not subjected to photoreactivation (see, for example, [12]).

Therefore, to obtain effective inactivation, a radiation source that provides minimal resistance to radiation for the entire range of bacterial infectious agents and, at the same time, has a relatively narrow radiation spectrum that excludes or minimizes photoreactivation is required.

Low-pressure mercury lamps (LPMLs) are traditionally used as a source of such optical radiation. In their spectrum, about 60% of the radiation flux falls on the fraction of the resonance emission line of mercury at a wavelength of 253.7 nm (Fig. 1, curve *1*). Although this emission line is slightly farther from the first maximum of the action spectrum (Fig. 1, curve *2*), it is near the long-wavelength (first) maximum of the integral absorption spectrum of DNA (Fig. 1, curve *3*) [13]. This provides an almost optimal inactivating effect on bacteria. Therefore, irradiators based on LPMLs are called bactericidal installations. However, a serious disadvantage of these lamps is the presence of mercury, which leads to increased risks of environmental pollution when the flask is depressurized and complicates the storage, operation, and disposal of such equipment. EU countries have been phasing out lamps containing mercury for more than 10 years. The same processes have begun in the Russian Federation.

3. EFFECTS OF EXCILAMP RADIATION ON BACTERIA AND LIVING CELLS

While studying excilamps in 2006, attention was given to the fact that the maximum radiation power of an excilamp based on XeBr* molecules (282 nm) is located approximately at the same distance from the maximum of the inactivating action spectrum as the LPML resonance line (Fig. 1, curve *4*). In addition, the emission spectrum of the XeBr excilamp has a short-wavelength "tail" in the wavelength range of 260–282 nm, which covers half of the first DNA absorption peak and the action spectrum [15]. It was assumed that both radiation sources (XeBr excilamp and LPML) should have the same bactericidal effect at the same energy exposures, and this was confirmed on the *Escherichia coli* test culture (ATCC 25922). Upon repeated inactivation of the surviving bacterial cells, it was found that they acquire resistance to LPML radiation, and the inactivating effect of the XeBr excilamp radiation does not change. This phenomenon was associated with the fact that the radiation spectrum of the excilamp is wider than that of the LPML. Accordingly, a comparatively large number of DNA defects can be expected upon exposure to an excilamp.

Next, a direct comparison was made of the bactericidal action of the LPML and XeBr excilamp on several bacterial strains, including *E. coli* (501), *Klebsiella pneumonia* (ATCC 2482), *S. aureus* (209P), and the clinical isolates of *Proteus vulgaris*, *Pseudomonas aeruginosa*, and *Candida albicans* [16]. The comparison showed that the XeBr excilamp is not inferior to the LPML in its bactericidal efficiency and, in some cases, surpasses it. As the XeBr excilamp does not contain mercury, is less sensitive to ambient temperature than the LPML, easily ignites, and instantly moves into operation, it can be concluded that the LPML has a serious alternative in terms of inactivating bacteria.

Another alternative to LPMLs can be LED radiation sources with a spectrum in the range of 250–300 nm, which have been developed in recent years. For example, LED modules from LG Innotek Co. (Republic of Korea) provide peak bandpass emissions at 266, 270, 275, and 279 nm with a half-width of about 15 nm. They are called UVC LEDs (UVC radiation refers to wavelengths of $200 < \lambda < 280$ nm). Figure 1 shows an example of the spectrum of one of the developed modules (curve *5*). In bacterial cultures of *Escherichia coli* O157:H7, *Salmonella enterica serovar Typhimurium*, and *Listeria monocytogenes*, the bactericidal effect of these sources was proved [14]. The advantage of these LEDs is their small size, which simplifies their arrangement in devices for disinfection. In addition, like XeBr excilamps, UVC LEDs do not contain mercury and do not require time to warm up when the device is started.

It can be assumed that UVC LEDs, which exhibit band radiation rather than linear radiation, will produce a greater number of DNA mutations in bacterial cells. This will prevent the emergence of resistance to radiation, similar to what was shown in [15] in relation to a XeBr excilamp. But to state this definitively, in the future, it is necessary to conduct comparative studies on the effect of radiation of LEDs and LPMLs on bacteria, which consider changes in the resistance of experimental bacterial cultures. In addition, separate studies on the issue of photoreactivation of common pathogens found in bacterial infections under the influence of such radiation sources are needed. Nevertheless, UVC LEDs should be classified as bactericidal radiation sources.

A separate, interesting issue is the effect of UVC radiation on living cells, which was studied in [17,18]. The object of research was represented by the living cells of the ovary of the Chinese field hamster *Chinese Hamster Ovary* (CHO-K1), which were exposed to radiation from a XeBr excilamp and an iodine vapor lamp at the 206 nm atomic line [19]. It was shown that the inactivating effect of UV radiation on a living cell differs significantly from the effect on a bacterium. The DNA of a living cell is slightly damaged due to direct exposure to UVC radiation. This damage is the result of a chain of reactions: 1) photon + substrate → radicals,

2) radicals + cell components (including DNA) → oxidation and inactivation of cell components. In addition, the cell produces antioxidants able to regulate the rate of their formation in its internal environment [18]. Therefore, a living cell is capable of resisting UV inactivation until a certain critical dose, after which its necrosis occurs. Hence, by selecting the dose of UVC irradiation, it was possible to carry out selective bacterial sterilization of wounds without inactivation of living cells of the body.

4. EFFECTS OF EXCILAMP RADIATION ON VIRUSES

Today, the most urgent area of research is the inactivation of viruses (the so-called virucidal effect of radiation). Bacteriophages are traditionally used as model objects that demonstrate the virucidal effect of radiation sources in these studies [20]. Thus, in [21], bacteriophage MS2 (strain VKPM RN-1505), which multiplies on a culture of *E. coli* K 12 *F*+ (strain VKPM B-3254), was used to compare the virucidal effect of LPML and XeBr excilamp radiation. The radiation from both sources effectively inactivated the bacteriophage, but its sensitivity to the action of the XeBr excilamp radiation was higher than that of the LPML. From an optical perspective, this can be explained by the fact that the radiation spectrum of the excilamp (Fig. 1, curve *4*) includes wavelengths that are absorbed by proteins that form the phage envelope and protect the genome, as well as the RNA of the bacteriophage.

A detailed study of the effect of UVC radiation in the 210–290 nm range on several bacteriophages was carried out in 2015 [22]. The optical characteristic of the radiation efficiency was the action spectrum, the function $S(\lambda)$, reflecting the reaction of bacteriophages to identical doses of UV radiation when exposed to radiation with different wavelengths. The S value (253.7) for the LPML ($\lambda = 253.7$ nm) was taken as a unit [23]. An example of the spectrum obtained for bacteriophage MS2 is shown in Fig. 2 (curve *1*). In this case, as in all others, spectra are characterized by a noticeable (i.e., manifold) increase in the sensitivity of phages to UV radiation at wavelengths shorter than

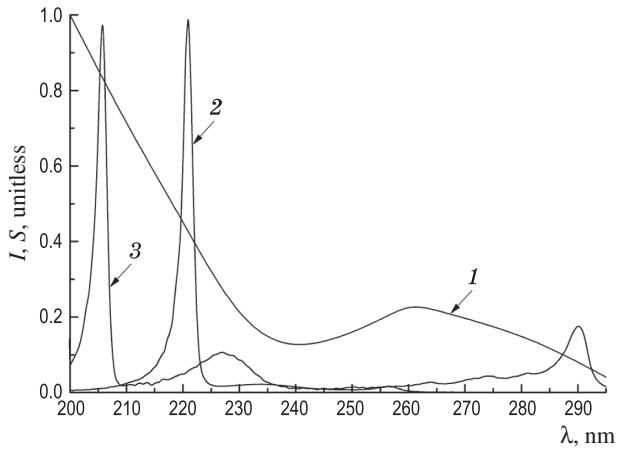

Fig. 2. *1*—action spectrum for inactivation of bacteriophage MS2 $S(\lambda)$, reconstructed according to the data of [22], *2*—emission spectrum of a KrBr barrier discharge excilamp, and *3*—emission spectrum of a KrCl barrier discharge excilamp.

240 nm as compared with 240–290 nm wavelengths. These data are consistent with the results [24,25].

Reasons for the increased sensitivity of phages to radiation in the range of $200 < \lambda < 240$ nm have yet to be established, although the facts known in photobiology are already sufficient to suggest the following explanation. Short-wave UV radiation damages not only the nucleic acid of the virus (DNA or RNA), but also the proteins that form the capsid (the envelope of the virus). These proteins not only protect the virus's nucleic acid from the external environment, but also ensure the attachment of the virus to the host cell. The amino acids that form these proteins absorb energy at $\lambda < 230$ nm, exceeding the absorption of total DNA. Therefore, damage to phage proteins by short-wave UVC radiation should lead to a decrease in the ability of viruses to infect host cells.

Accordingly, the best inactivation of viruses (in comparison with LPMLs) would be provided by UVC radiation sources, the spectrum of which is concentrated in the wavelength range of $200 < \lambda < 240$ nm. Based on this, the concept of radiation sources of specialized virucidal action has been introduced and circulated. For irradiation in the indicated wavelength range, barrier-discharge excilamps based on $KrBr^*$ and $KrCl^*$ molecules are suitable, the emission spectra of which are shown in Fig. 2 (curves *2* and *3*). Both spectra have an intense $B \rightarrow X$ emission band with a maximum at 207 and 222 nm and a half-width of 2.18 and 2.04 nm, respectively.

Various versions of the design of excilamp-based irradiators are possible. The excilamp can be housed in a structure equipped with a reflector and power supply, such as the portable model shown in Fig. 3 (*1*). Such models have a relatively small size and weight of the output window (60 × 90 mm, 0.7–0.8 kg) and provide an energy luminosity of up to 30, 20, and 10 mW/cm^2 for working molecules $XeBr^*$ (282 nm), $KrCl^*$ (222 nm), and $KrBr^*$ (206 nm), respectively. The model can be scaled and used as an open-type irradiator for disinfection of surfaces and indoor air. Figure 3 (*2*) shows another version, in which the excilamp and power supply are made separately. In this model, the KrCl excilamp has an emitting surface length of 35 cm with a diameter of 4.3 cm and provides an energy luminosity of 13 mW/cm^2.

As noted earlier, short-wave UVC radiation also meets the need for selective treatment of wounds and skin, when

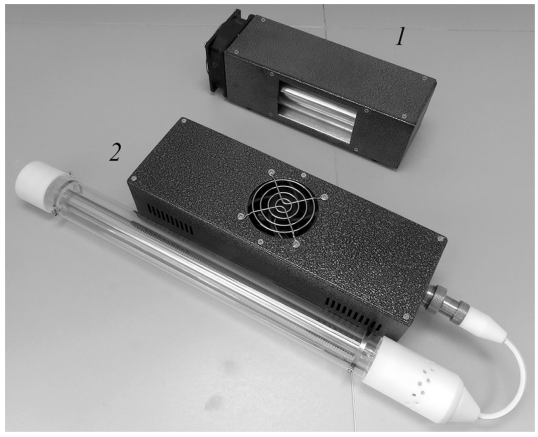

Fig. 3. Designs of irradiators developed at IHCE SB RAS with internal (*1*) and external (*2*) excilamps.

inactivation of infectious agents is required without harmful (i.e., dose-limited) effects on living cells of the human body. It is known that the depth of UV radiation penetration into living tissues of mammals decreases nonlinearly with decreasing radiation wavelength [26]. Therefore, damage to living cells from virucidal radiation from excilamps should be lower than that of LPML radiation. These considerations were confirmed experimentally by using the KrCl and KrBr excilamps we created; the model is shown in Fig. 3 (*1*) in the work of the Center for Radiological Research (New York, USA) [27,28]. Specifically, it was shown that there is a narrow wavelength band in the short-wavelength part of UVC radiation (approximately 200–222 nm), which inactivates bacteria without damaging tissue cells. Comparison with LPMLs showed that radiation from excilamps causes hardly any DNA damage and is not cytotoxic for the open skin of mammals. Similar confirmations were obtained in [29,30].

Moreover, in 2018, it was experimentally confirmed that radiation from a KrCl excilamp (model *1* in Fig. 3) at doses of 2 mJ/cm^2 inactivates more than 95% of the H1N1 influenza virus in the form of an aerosol [31].

In connection with the spread of coronavirus infections, interest in studies of the virucidal effect of KrCl excilamps has increased. The first study [32] concerned the inactivation of two harmless human coronaviruses, alpha HCoV-229E and beta HCoV-OC43. It has been estimated that, with continuous exposure to a KrCl excilamp in public places at the currently recommended exposure limit (3 $mJ/cm^2/h$), 99.9% viral inactivation can be achieved within approximately 25 minutes for the HCoV-OC43 beta coronavirus. Since all coronaviruses have a similar structure and the same RNA chain length, it can be deduced that inactivation of other coronaviruses, including SARS-CoV-2, by KrCl excilamps, will be similarly efficient.

At the present time, many experimental confirmations that support these conclusions have been obtained. In [33], the virucidal effect of a KrCl excilamp on surfaces infected with SARS-CoV-2 was studied in vitro. At irradiation doses of 1 and 3 mJ/cm^2, the inactivation efficiency was 88.5% and 99.7%, respectively. However, the authors note that their laboratory experience does not negate the need for research on decontamination of real surfaces.

In [34], the absorption spectra of solutions containing SARS-CoV-2 virus particles at a concentration of 10^5 CFU/mL were obtained for the wavelength range of 200–300 nm. The measurements showed that radiation from the KrCl excilamp was absorbed by the solution approximately 3.5–4 times stronger than that of LPML radiation.

In [35], UVC radiation acted on bacteriophage Phi6 and two coronaviruses—human coronavirus (HCoV) 229E and murine hepatitis virus (MHV). The sources of UVC radiation were a KrCl lamp, an LPML, and two tabletop UV LED systems with peak radiation wavelengths of 270 and 282 nm. Comparative tests have shown that among the sources of UVC radiation, the KrCl excilamp provides the best virucidal effect for all three enveloped viruses.

The authors of [36], summarizing the literature data on the virucidal effect of various wavelengths (from ultraviolet to infrared), concluded that, to achieve 63% inactivation in the case of a KrCl excilamp, energy exposures are required that are orders of magnitude lower than for LPMLs and other long-wave radiation sources. It is noted that short-wave UVC radiation provides "adequate virucidal doses in a reasonable time, in contrast to other wavelengths that require higher doses and longer exposure times" [36].

5. TRENDS IN THE DEVELOPMENT OF IRRADIATORS BASED ON EXCILAMPS

Irradiators based on UVC excilamps show all the signs of a promising technology for disinfecting air and surfaces from infectious agents of various etiologies. As such, it can be predicted that the further development of the nascent industry will take place in the following three directions:

1) obtaining data on the inactivating effect of UVC radiation on new microbiological objects, including those in different environmental conditions, on various real surfaces, and in water (e.g., [37], where the effect of radiation on spores and/or growing cells of bacteria of the genera *Bacillus* and *Clostridium*);
2) revealing the effect of long-term use of UVC radiation from excilamps on human health and the degradation of materials on irradiated surfaces (the need for this was noted in [36]);
3) creation of a family of irradiation installations based on UVC excilamps and their technological improvement.

Specifically, based on the developments of the IHCE SB RAS, an air recirculator based on a KrCl excilamp was released in 2020 by the Joint Stock Company "Research and Development Centre 'Polyus.'" The device was designed for air disinfection in the presence of people and was distinguished by its relatively small dimensions, modern circuitry solutions, and layout. In the same year, the device received a conclusion from the Federal Budgetary Institution of Science State Research Center of Virology and Biotechnology "VEKTOR" of Rospotrebnadzor that it was recommended for widespread use in disinfecting air from viral and bacterial aerosols in household premises and microbiological laboratories.

It is known that sources of short-wave UVC radiation produce ozone in the ambient air due to the direct action of radiation on the air and due to processes on the high-voltage electrodes of the lamp. The first pathway of ozone formation is easily compensated for by air circulation in the room. To compensate for the action of the second path, an irradiator and air recirculator, ORVIK-1/222, was developed at the IHCE SB RAS in 2021. The device can operate both in the open irradiator mode, for which a removable casing is provided (Fig. 4), and in the closed air recirculator mode, providing an energy luminosity of up to 5 mW/cm^2 with a lamp length of 27 cm. The levels of ozone production are decreased by placing the excilamp in a cover that is transparent to UV radiation [38]. Without it, the above-described excilamp gives an excess of the maximum permissible concentration for ozone in a closed room, with a volume of 40 m^3 after only 30 min. This does not happen in the ORVIK-1/222 model, enabling the unit to be used continuously.

One more potential line of development for irradiators should be mentioned. Due to the variety of infectious agents,

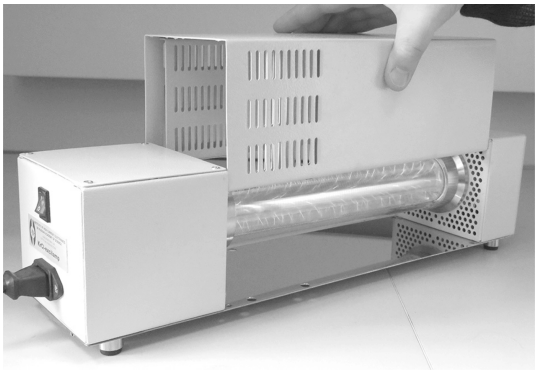

Fig. 4. Design of the ORVIK-1/222 irradiator and air recirculator developed at the IHCE SB RAS.

it is necessary to create universal devices for disinfection that reduce both bacterial and viral contamination of volumes and surfaces. To address this problem, it is likely to predict the appearance of devices in which, in addition to a virucidal excilamp emitting in the wavelength range of 200–240 nm and generating radiation locally, bactericidal radiation sources (LPMLs, UVC LEDs, XeBr excilamps) will be installed. As a result, this approach should improve the quality of processing for normal conditions, when a given volume or surface to be treated contains not only bacteria, but also viruses. Moreover, the use of narrow-band and line radiation sources in this task is likely to reduce both the proportion of photoreactivation and the proportion of resistant mutations in microorganisms emerging.

6. CONCLUSIONS

The results presented in this study indicate that there are differences between the action of short-wave ultraviolet radiation on viral and bacterial particles. These differences are reflected in the disparity of their spectral characteristics, which ultimately lead to variations in the inactivation of these particles. These variations are the fundamental basis for the development of new virucidal technologies, as well as the combined action of virucidal and bactericidal technologies, a necessary element of optical radiation sources based on $KrCl^*$ and $KrBr^*$ molecules.

Acknowledgment. The authors express their deep gratitude to their colleagues in the Department of Microbiology and Virology of the Siberian State Medical University (Russia, Tomsk), the Department of Cytology and Genetics of Tomsk State University (Russia, Tomsk) and the Faculty of Biomedical Engineering of the University of Eindhoven (the Netherlands, Eindhoven) for their assistance in studying the effect of the ultraviolet radiation of excilamps on microorganisms. This article was prepared within the framework of the State Assignment of the IHCE SB RAS, project no. FWRM-2021-0014.

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