Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

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Review

The inactivation and destruction of viruses by reactive oxygen species generated through physical and cold atmospheric plasma techniques: Current status and perspectives

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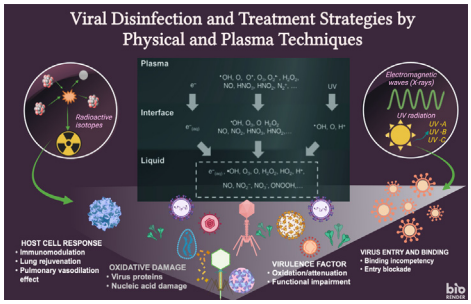
HIGHLIGHTS

- Reactive oxygen species (ROS) can be employed in assisting host defense against viruses.
- ROS-based strategies provide application for effective sterilization and disinfection.
- Cold plasma delivery system as an eco-friendly tool for virus destruction.
- These techniques can kill viruses by their surface modification to prevent host infection.
- ROS as therapeutic mediators against infection is a promising avenue for treatment.

GRAPHICAL ABSTRACT



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ABSTRACT

Background: Outbreaks of airborne viral infections, such as COVID-19, can cause panic regarding other severe respiratory syndrome diseases that may develop and affect public health. It is therefore necessary to develop control methods that offer protection against such viruses.

Aim of Review: To identify a feasible solution for virus deactivation, we critically reviewed methods of generating reactive oxygen species (ROS), which can attack a wide range of molecular targets to induce antiviral activity, accounting for their flexibility in facilitating host defense mechanisms against a comprehensive range of pathogens. Recently, the role of ROS in microbial decontamination has been critically investigated as a major topic in infectious diseases. ROS can eradicate pathogens directly by inducing oxidative stress or indirectly by promoting pathogen removal through numerous non-oxidative mechanisms, including autophagy, T-cell responses, and pattern recognition receptor signaling.

Key scientific concepts of review: In this article, we reviewed possible methods for the *in vitro* generation

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of ROS with antiviral activity. Furthermore, we discuss, in detail, the novel and environmentally friendly cold plasma delivery system in the destruction of viruses. This review highlights the potential of ROS as therapeutic mediators to modernize current techniques and improvement on the efficiency of inactivating SARS-CoV2 and other viruses.

Introduction

Diseases and health conditions caused by microorganisms have always been a challenge in the medical sciences. Bacteria, fungi, and viruses can invade human hosts and disrupt the normal physiological balance by producing toxic by-products. Viruses are intracellular pathogens that contain either DNA or RNA as genetic material [1] and virus-borne diseases are currently one of the most serious concerns faced in medical sciences. For centuries, viral diseases have been responsible for several deaths and in the eighteenth century, the poxvirus killed over 100 million people in Europe [2]. Similarly, in 1901, 1918, and 2014, yellow fever, the Spanish flu, and the Ebola outbreak caused by the flavivirus [3], the H1N1 [4], and the Ebola viruses, respectively, killed millions of people worldwide [5,6]. Recently, in 2019, the novel coronavirus originated in Wuhan, China [7] and was declared a global pandemic according to the World Health Organization (WHO) [8,9]. The main drawback of viral infections is that the virus can hijack the cellular machinery of the host and nucleic acid (DNA and RNA)-based viruses have advanced defense mechanisms for escaping host recognition [10]. As viruses are pathogens with a comparatively rapid mutative rate, particularly RNA viruses, virus–host co-evolution relies on rapid identification, the host's immune system response, and virus-employed evasion mechanisms, resulting in a constant interaction between escape/spread and immunity/-clearance. To that effect, there are various mechanisms by which the host senses and responds to viral infections [11,12].

Over the last few decades, large-scale vaccination programs have been conducted to combat viral infections, such as polio [13], smallpox [14], and hepatitis [15]. These vaccinations have been the most successful strategy for preventing these deadly viral diseases to date [16,17]. However, biochemical agents that are effective against viral infections have been used to develop antiviral medications. Biochemical antiviral therapeutics are mainly used for common viral infections and are popular as alternative treatment approaches. Apart from vaccination, other preventive measures are also important in protection against viral diseases, such as social distancing and quarantining when exposed, as isolation is a basic step that minimizes the spread of viruses in a community. To inactivate or kill viruses in certain environments or reduce viral spread among people, other strategies have been invented and implemented. Disinfectants, organic and inorganic chemicals, and nanomaterials are used to inactivate viruses, whereas alcohol, different types of surfactants, and oxidizing agents are widely used as sterilizing agents against viruses [18]. Recently, the application of different metal nanoparticles for virus inactivation has gained attention [19]. In addition, physical methods, such as radiation, lasers, photodynamic therapy (PDT), and non-thermal atmospheric plasma, have been used for disinfection in industrial environments (Fig. 1). These "virucidal" chemicals and techniques can destroy viral cells and modify their surface structures to prevent them from infecting potential host cells.

In this review, we summarize the currently available and commonly used methods for disinfection and the possible application of new strategies to efficiently inactivate viruses in the environment using reactive oxygen nitrogen species-based techniques [20]. Currently, due to the COVID-19 pandemic, it is imperative to find an effective sterilization technique that significantly reduces the spread of viruses. To that effect, the potential of cold plasma technology has been discussed for effective virus deactivation in future applications.

Reactive oxygen species (ROS) and their role in anti-viral responses

Metabolites derived from nitric oxide (NO•) and superoxide (O_2•−) play significant roles in antiviral defense; however, they may also damage the host. In fact, certain levels of these metabolites may aid viral replication because of their mitogenic effects on cells [21]. Cellular ROS generation is often induced by both exogenous and endogenous stimuli, as ROS act as key cell-signaling molecules for normal biological development. However, ROS formation may also damage many cellular organelles and processes, ultimately interrupting the normal physiology of cells. An essential concept to keep in mind when assessing the oxidative stress levels of a cell is whether an elevated oxidant status prompts biomolecule damage and determines the threshold vital to cellular processes via redox signaling [22]. ROS can be generated by various drugs and conventional physical means, such as X-rays and gamma radiation, including PDT. As these methods have been extensively investigated, this review focuses more on cold plasma-induced ROS generation mechanisms.

In cold plasma, a wide variety of reactive species are formed, based on numerous parameters, such as the feeding gas, configuration of the target material, energy source, and distance between the target and plasma source during plasma discharges. A schematic diagram of the reactive species produced within the discharge area and the plasma–liquid interface is displayed in Fig. 2.

Primary reactive species, such as nitric oxide radicals (NO), excited nitrogen (N), hydroxyl radicals (OH), superoxide radicals (O_2^{-*}), and singlet oxygen ($^1O_2$), are directly generated in the plasma discharge zone because of the interaction between free electrons and the feeding gas molecules. These species have a limited lifetime; however, their concentration in the discharge area is extremely high. More importantly, several studies show that the intensity of the OH radicals produced in ambient air is extreme [23 24 25]. The species are further converted to prolonged reactive species, such as hydrogen peroxide (H_2O_2), nitrite (NO_3), ozone, and nitrate (NO_2), in the ambient atmosphere. The primary reactive species may react with ambient air molecules, eventually producing stable species. Among these, H_2O_2 is very dispersible and stable inside water, whereas NO_3 and NO_2 are transformed to NO_3^- and NO_2^-, respectively. It is worth mentioning that the chemistry behind reactive oxygen and nitrogen species [20] generation varies widely depending on different plasma discharge conditions. A short description of the main reactive species produced in plasma discharges is provided below.

Atomic oxygen

Atomic oxygen (O) is a monatomic oxygen that is highly reactive and can rapidly bond with surrounding molecules. In atmospheric-pressure plasma, atomic oxygen can be generated

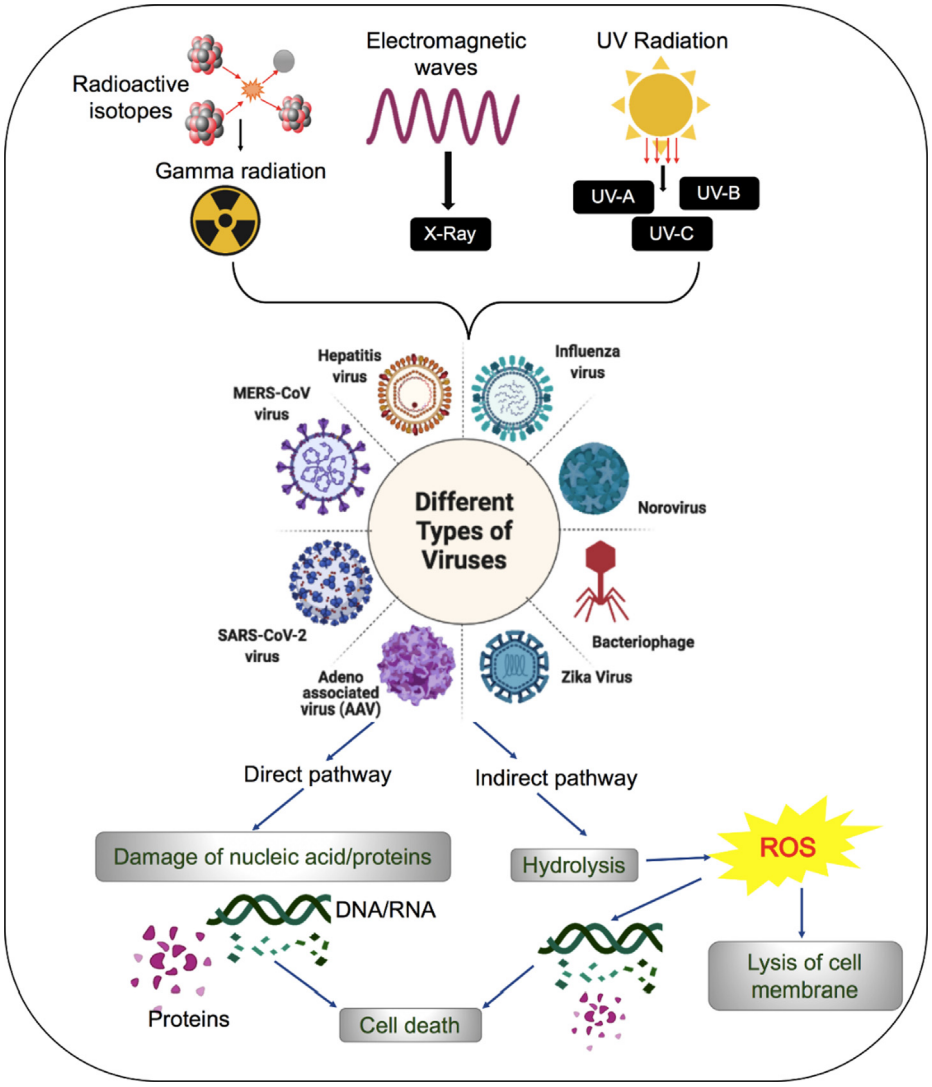

Fig. 1. Direct and indirect damage of microorganisms by different types of radiation. Image was created using Biorender.

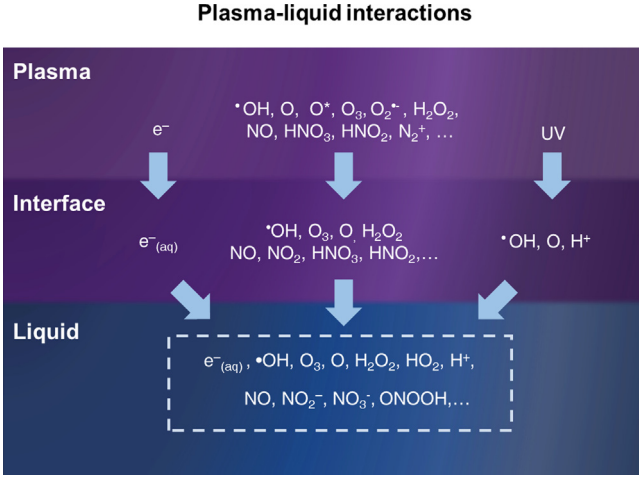

Fig. 2. Schematic indicating the generation of different reactive species in the plasma, plasma-liquid interface, and liquid phase under plasma irradiation.

by the collision of O_2 molecules with metastable plasma working gas (M^*) and excimers (M_2^*) [26].

$$M^* + O_2 \rightarrow M + 2O \text{ (M: He, Ne, Ar.)}$$

$$M_2^* + O_2 \rightarrow 2\ M + 2O$$

Atomic oxygen can also be formed through the electron-impact dissociation of the O_2 molecule [27].

$$e^- + O_2 \rightarrow O + O + e^-$$

It exhibits a strong emission at 777 nm, which can be easily observed using a simple optical emission spectrometer.

Hydroxyl radical

The hydroxyl radical (•OH) is a strong oxidizing agent and one of the main reactive oxygen species that contributes to–the plasma-liquid interactions. In a plasma system, •OH radicals are formed by the dissociation of water molecules in both the ambient environment and liquid phase [26].

$$M^* + H_2O \rightarrow M + H^\bullet + {}^\bullet OH \text{ (M: He, Ne, Ar.)}$$

$$M_2^* + H_2O \rightarrow 2\ M + H^\bullet + {}^\bullet OH$$

$e^- + H_2O \rightarrow {}^\bullet OH + H + e^-$

The plasma-induced ultra-violet water photolysis process with an energy of 4–6 eV can also generate $^\bullet$OH in bulk liquid [25].

$UV + H_2O \rightarrow H_2O^*$

$UV + H_2O^* \rightarrow H^\bullet + {}^\bullet OH$

$H_2O^* \rightarrow OH^- + H^+$

$OH^- \rightarrow {}^\bullet OH + e^-$

Other important pathways of $^\bullet$OH generation are as follows [28]:

$e^- + O_2 \rightarrow O(^3P) + O(^1D)$

$O(^1D) + H_2O \rightarrow 2\ {}^\bullet OH$

$NO + HO_2 \rightarrow {}^\bullet OH + NO_2$

$O_2 + H \rightarrow {}^\bullet OH + O$

The hydroxyl radical is one of the most common ROS in cold atmospheric-pressure plasma systems. In addition to its high chemical reactivity, $^\bullet$OH can provide important physical parameters for plasma. It exhibits an emission signature at 309 nm, which can be used to calculate the gas temperature of the plasma system [29].

Superoxide/Hydrogen peroxide

Superoxide (O_2^-) and its protonated form, hydrogen superoxide (HO_2), play important roles in microbial cell inactivation and related cellular processes. Superoxide radicals can be produced by plasma through electron-impact collisions [27].

$e^- + O_2 \rightarrow O_2^-$

Superoxide is also generated by the reaction between plasma-generated $^\bullet$OH and ozone [30].

$^\bullet OH + O_3 \rightarrow HO_2 + O_2$

In aqueous environments, O_2^- and HO_2 are in chemical equilibrium:

$O_2^- + H_2O \leftrightarrow HO_2 + OH^-$

The concentration of O_2^-/HO_2 in plasma is relatively lower than that of other dominant ROS products, yet they are important factors in biomedical applications of plasma systems [27].

It is believed that H_2O_2 is produced in cells under normal and stressful conditions, such as UV radiation, chilling, wounding, and infection. Owing to the fact that H_2O_2 does not have any unpaired electrons, it may freely cross cellular membranes and subsequently induce oxidation. Bienert *et al.* [31] suggested that H_2O_2 is a single ROS that may diffuse through aquaporin inside the cellular membrane and is more stable than other ROS. Therefore, ROS act as signal molecules that help regulate particular biological developments, increasing tolerance to several environmental stresses [32]. As mentioned, H_2O_2 is a stable molecule that is formed under humid conditions owing to the combination of OH radicals. Examples of H_2O_2 and OH reaction mechanisms are as follows:

$OH + OH \rightarrow H_2O_2$

$H_2O_2 + OH \rightarrow HO_2 + H_2O$

In addition to direct plasma discharge to target samples, plasma-activated medium (PAM) can be generated by exposing a biological liquid to electrical discharge under atmospheric conditions. When plasma is produced in ambient atmosphere, it generates a large concentration of reactive species. These RONS are important species that make the cold plasma technique applicable in numerous biomedical fields. For plasma-related biomedical applications, a certain target can demand off-site or on-site exposure conditions. In on-site conditioned treatments, the active plasma plume relatively close to the ambient environment comes in direct contact with biological objects. This method could be valuable for treating critically affected wounds or contagious decontamination. In the off-site method, plasma is directly exposed to biological solutions (supplemented with a diverse range of RONS), which are utilized for specific biomedical applications. Interestingly, these types of plasma-exposed solutions can also be applied to areas where there is no option for plasma production. They can also serve as therapeutic drugs and are termed as plasma-activated solutions (PAS). Notably, the levels of the reactive species generated can be controlled by varying the feeding working gas, flow rate, and applied voltage. Short-lived reactive species can be employed in *in-situ* treatment settings, whereas long-lived reactive species can be preserved in PAM for further use. Plasma technology is straightforward and affordable, enabling its application in many fields.

Ozone

Ozone is an allotrope of oxygen that has strong oxidative activity with a distinctive smell. Under ambient conditions, O_3 is unstable and readily decays into molecular oxygen. It is considered a dangerous gas for human health. In atmospheric-pressure plasma, O_3 can be generated by a third-body collision between O and O_2 in the presence of a third body molecule (M') [27].

$O + O_2 + M' \rightarrow M' + O_3$

Even with low stability, O_3 can exist for several minutes to hours in an ambient environment; thus, controlling the generated O_3 concentration by the plasma sources within the safety condition is necessary. However, O_3 shows low water solubility, which makes it less important than other ROS in plasma-liquid interactions.

Methods for viral deactivation

Existing physical techniques to eliminate viruses

Ultra-violet radiation

Several physical methods have been adopted for the decontamination of different viral species, including irradiation, X-rays, and PDT (Table. 1). Irradiation offers a low-energy, environmentally friendly, and safe method of killing viruses under well-controlled circumstances with the advantage of minimal molecular changes, which is especially important in the preparation of biological reagents. Two distinct advantages of irradiation over other methods are the linearity of the lethal dose effects and the ability to measure the dose to be delivered [33]. The key natural germicide is the sun-produced UV radiation in the atmosphere. In contrast, far-UVC light (207–222 nm) proficiently eradicates pathogens without affecting their normal counterparts [34]. Nucleic acids within viral particles play a vital role in UV absorption and viral inactivation. At a range of 254 nm, UV radiation inactivates and tabulates the susceptibilities of a wide range of viruses, including those with double/single-stranded RNA or double-stranded DNA genomes [35]. Two issues must be considered when solar inacti-

Table 1
A list of well-established physical methods for various virus decontamination.

Types of radiation	Decontaminating virus	Dose	Source	Reference
		207–222 nm	Far-UVC light	[34]
		broad (200–320)	UV	[74]
		254-nm	UV	[75]
		254-nm	UV	[76]
		254 nm	UVC photon	[77]
		280–315 nm	UV-B	[36]
	SARS-CoV-2	280 ± 5 nm	UVC	[78]
		PX-UV robot	PX-UV	[79]
		model PXUV4D		
		207–222 nm	Far-UVC light	
	HCoV-OC43	222 nm or 3 log10	UV	[80]
	HCoV-229E			
	Variety of virus	254-nm	UV	[35]
	Live influenza A (H1N1)	207–222 nm	Far -UVC light	[81]
Ultraviolet radiation	ARS-CoV-2	222-nm or 0.94 log10	UVC light	[82]
	SARS-CoV, CCHFV or NiV	0.2 J/cm2	UVC light	[83]
	Animal virus (VSV) and Bacteriophage (MS2)	2250–3020 Å	UV	[37]
	Herpes simplex virus	254-nm	UV	[84]
	MERS CoV		UVC light	[85]
	SARS CoV-1	254 nm	UVC	[86]
	Polyoma virus	100 erg mm-* second^{-1}.	voltage-stabilized low pressure mercury vapor lamp.	[51]
	MNV-1			
	VLPs	4 kGy	Cobalt-60	[49]
	VSV		Cesium 137	
	Calicivirus	253.7-nm	UV	[87]
	CoV-1 and CoV-2	1×10^6 $TCID_{50}$/mL	Cobalt-60	[44]
	SARS-CoV-2	1.0 Megarad	Cobalt-60	[61]
	SARS-CoV-1			
	Coronavirus	150 Gy	Co-60	[62]
	HIV	2.5 J M^{-2} S^{-1}	UV	[46]
			Co-60	
Gamma radiation	BSL-4 virus	$TCID_{50}$/mL	Cobalt-60	[42]
	LACV	0.5 Mrad		
	Animal virus	93 ergs/g	Cobalt-60	[88]
	Polyoma virus	No dose rate	Cobalt-60	[51]
	Vaccinia virus	37 nm	Cobalt-60	[53]
	Newcastle disease virus			
	Influenza virus			
		No dose rate	Cobalt-60	[54,55 36]
	Arenavirus			
	Bunyavirus			
	Coronavirus	$TCID_{50}$/mL	Cobalt-60	[60]
	Filovirus	1–5 MRad		
	Flavivirus			
	Orthomyxovirus			
	Paramyxovirus			
	Akabane virus	20–0.55 Megarads	Cobalt-60	[89]
	Bunyaviridae			
	Reoviridae			
	Parvoviridae			
	Coronaviridae			
	Newcastle disease virus			
	Ainoa	0.35 megarads	Cobalt-60	[90]
	Akabanea	0.25 megarads	Cobalt-60	[90]
	BEFVa	0.29 megarads	Cobalt-60	[90]
	Hog cholera	0.55 megarads	Cobalt-60	[90]
	SARS-CoV-2	1 Megarads	Cobalt-60	[91]
Photodynamic therapy	Polio virus	No dose rate	Toluidine blue	[63]
	Herpesvirus	0.5 μg/mL	Hematoporphyrin	[92]
	vesicular stomatitis virus		Pristine C60	[93]
X-Ray	Zoonotic viruses (laviviridae, Nairoviridae, Phenuiviridae and Togaviridae)	50 kGy	X-ray Irradiator	[94]
	Influenza virus	90,000 roentgens per minute (r/min).	Machlett OEG-60	[72]
	Polyoma virus	14.4–48 kiloroentgens per minute.	Holweck X-ray tube	[51]

SARS-CoV-2: Severe acute respiratory syndrome coronavirus 2; HCoV-OC43: Human coronavirus OC43; HCoV-229E: Human coronavirus 229E; CCHFV: Crimean-Congo haemorrhagic fever virus; NiV: Nipah virus; VSV: Vesicular stomatitis virus; HSV: Herpes simplex virus; MERS CoV: Middle east respiratory syndrome coronavirus; SARS CoV-1, Severe acute respiratory syndrome coronavirus 1; MNV1: Murine norovirus 1; VLPs: Virus-like particles; CoV-1: Coronavirus 1; CoV-2: Coronavirus 2; HIV: Human immunodeficiency virus; BSL-4: Biosafety level 4; LACV: La Crosse virus; NDV: Newcastle disease virus; BEFV; Bovine ephemeral fever virus.

vating bio-threat viruses: 1) assessing the UV sensitivity of different viruses and 2) knowing the number of RNA or DNA bases to determine the sensitivity to UV inactivation at a specified UV [36]. Additionally, significant variance in susceptibility among viral genome types occurs because of pyrimidine dimers, particularly thymine dimers, which are the most naturally toxic UV photoproducts. As DNA contains thymine, DNA viruses are highly susceptible to UV damage compared to RNA viruses [37]. Furthermore, the deadly effect of UV could be suppressed by the repair, particularly for double-stranded genomes containing viral structures [38]. The most effective generator of UV beams for microbial applications is a xenon and mercury-vapor arc lamp [39]. A recent study showed that SARS-CoV2 can be effectively inactivated by UVC irradiation. Recent studies have also highlighted the application of UV radiation for decontaminating N95 respirators to ensure safety against COVID-19 [40].

Gamma radiation

Gamma radiation is a type of ionizing radiation that can inactivate both DNA and RNA viruses [41,42]. Gamma rays have the smallest wavelength and greatest energy of the rays in the electromagnetic spectrum [43]. The basic mechanism underlying the inactivation of viruses through irradiation is thought to be the breakdown of nucleic acids (DNA/RNA) by radiolysis or genetic material cross-linking [44]. In other words, it can either directly break down the DNA helix or create free radicals that damage the DNA structure [45]. Gamma irradiation is an effective method for sterilizing pathogenic organisms in the environment [46]. Owing to its strong decontamination capability, gamma radiation is widely applied in the sterilization of medical devices, injectable products, and food samples [47,48]. Gamma radiation for clinical use is mainly generated from radioactive isotopes, such as cobalt-60 and cesium-137, which are also established sources in the biocontainment field. However, it can penetrate food, and thus, its application in food decontamination is regulated very carefully. The highest dose permitted by the Food and Drug Administration for the treatment of fresh produce is 4 kGy. This radiation dose has been found to be successful for bacterial inactivation; however, it appears to be insufficient for norovirus, which requires a higher dose [49]. Many studies have showed the efficiency of gamma radiation to inactivate pathogenic viruses in laboratory environments [42,50]. Gamma radiation sterilization studies of animal viruses include polyoma virus [51], Rous sarcoma [52], vaccinia [53], Newcastle disease [54], influenza [55], smallpox [56], herpes simplex [57], EBOV, and hepatitis A viruses [58].

Owing to the current worldwide COVID-19 pandemic, many researchers have analyzed the efficiency of gamma radiation in inactivating this virus. Gamma radiation has been reported to play an important role in vaccine preparation via viral inactivation, as it has already shown potential for inactivation efficacy against other enveloped viruses [59]. In addition, specimens of two other members of the COVID family, CoV-1 and CoV-2, are inactivated by gamma irradiation [60,61]. During this pandemic, personal protective equipment (PPE) and respirators need to be sterilized before reuse and gamma radiation has been found to be an effective disinfectant [62].

Photodynamic therapy

Photodynamic therapy is a comparatively recent methodology for the inactivation of native microbes, such as viruses. It uses well-known dyes as functional agents [63]. The PDT effect is perceived whenever an appropriate photosensitizer and O_2 are exposed to light; hence, PDT is a local treatment limited to specific infection sites [64]. Once exposed to visible light, photosensitizer compounds react with O_2 to produce ROS, including singlet oxygen [65]. These reactive species damage proteins, nucleic acids, and lipids, ultimately leading to cell death. A few studies use well-known photosensitizing molecules for PDT, including chlorophyll, heme, and bacteriochlorophyll [66].

The genetic material of the viruses can be disrupted by PDT as photosensitizers may bind or intercalate with viral nucleic acids. Cationic complexes, such as methylene blue, can penetrate the outer layer of viral structures and eventually be introduced into the genome [67]. Viral proteins naturally endure structural adaptations, such as protein cross-linking. In particular, photooxidative impairment occurs in oxidation-prone amino acids. Moreover, the interaction of a photosensitizer with viral proteins can affect protein folding and consequently disrupt viral function [65,68,69].

X-Rays

X-ray irradiation is an established method for decontamination and food sterilization. High energy can be generated when an electron ray is employed on a metal foil [70]. X-ray irradiation is a very efficient alternative to thermal or chemical decontamination methods. It is a type of ionizing radiation that generates highly penetrative photons and can effectively inactivate viral species of different families, such as *Flaviviridae*, *Nairoviridae*, *Phenuiviridae*, and *Togaviridae* [71]. A low voltage of +10 in an X-ray machine using a Machlett OEG-60 tube can reduce the survival rate of the influenza virus [72]. A study conducted in 1943 reported that virus-induced papillomas in rabbits regressed following exposure to X-ray irradiation. The study also concluded that X-rays can reduce regression of the virus by blocking cellular division and initiating pathological changes that ultimately kill the virus [73]. For SARS-CoV2, ionizing radiation has become a very effective way to sterilize gloves, surgical masks, PPE, etc. [62].

Advanced cold plasma technique for virus deactivation

To date, the effectiveness of different plasma devices [95] and PAS [96,97] has been well documented [96,98,99]. They have been used for decontamination in the food industry, packaging, pharmaceuticals, and medical field. Cold plasma is an ionized gas containing a wide range of reactive species that can kill microorganisms (Fig. 3). In the last few centuries, plasma has been frequently recognized as an effective antimicrobial agent against multidrug-resistant microorganisms on non-living surfaces or contaminated and septic tissues. Therefore, it has become a promising medical device, but with several clinical issues. In the case of anti-viral activities, most studies have shown that plasma-induced ROS production is the main cause of viral inactivation. Studies on the effects of plasma on virus particles followed a favorable virus inactivation technique to reduce contamination and sterilization [100]. Here, we review the effects of different plasma devices on many viral species and virus-associated syndromes [101].

Plasma application against other viruses

Plasma has been applied to bacteriophages, DNA and RNA viruses. To analyze the mechanism of the impact of plasma application in viruses, a study was conducted in 2018 which analyzed both dielectric barrier discharge (DBD) devices and PAS on bacteriophages. It was found that the reactive species generated through plasma application eventually damaged DNA and proteins to disrupt virus cells [102]. Yasuda et al. [103] developed a biological test that could estimate the *in vivo* DNA damage of bacteriophage lambda viruses exposed to air plasma. Regarding air sterilization against airborne bacteriophages, Xia et al. proposed and fabricated a packed-bed DBD plasma reactor that efficiently deactivates MS2 bacteriophages in aerosols with an air flow speed of 170 L/min using a nominal pressure drip through the reactor. This strategy is advantageous for protection against airborne viral diseases [104]. Furthermore, the inactivation of MS2 viruses in both water-

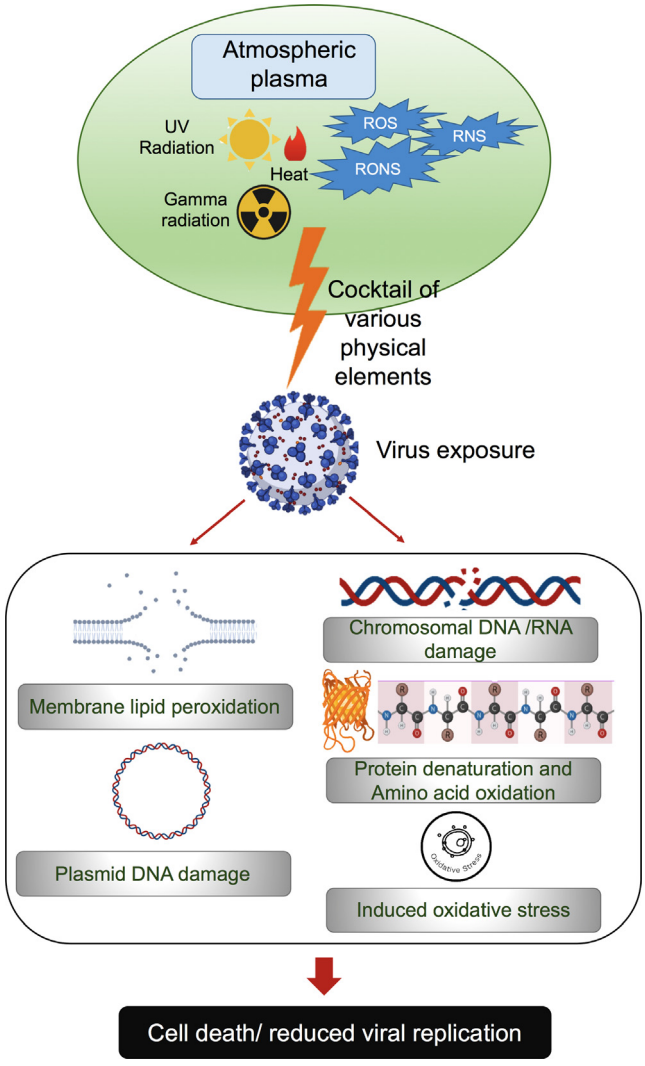

Fig. 3. Mechanisms of cold plasma-generated reactive species with respect to biological challenges in microorganisms. This image has been created using Biorender.

borne and airborne states largely relies on treatment time, given power, and feeding gas carrier type. This plasma-induced viral inactivation process is predominantly associated with the release of ROS, which eventually damages viral RNA and surface proteins [105]. Though these studies provide evidence that virus deactivation can be achieved by various cold plasma source applications, the primary mechanism by which inactivation occurs is not well understood. To elucidate this, Tanaka *et al* [106] performed plasma virus deactivations under both wet and dry conditions, and more damage was observed on the bacteriophage φX174 viral coat proteins than on DNA. Overall, plasma application renders bacteriophages inactive through ROS action. However, further investigation is required to understand the plasma operating conditions that favor protein inactivation or viral DNA damage.

In addition to bacteriophages, the inactivation of human respiratory viruses by cold plasma has recently been explored [107]. For instance, nitrogen gas plasma inactivates respiratory viruses, such as influenza, by damaging proteins and RNA [108]. Oxidation may be the most prominent factor in the degradation, inactivation, and modification of influenza A and B, including hemagglutinin, nucleoprotein, and neuraminidase viruses through N_2 gas plasma [109]. In adenoviruses, plasma has a virus-type-dependent effect, and surprisingly, infectivity could be amplified for some adenovirus types. However, the exact mechanism underlying this phenomenon remains unknown. Bunz *et al* [110] suggests that plasma treatment may be an effective antiviral therapy for certain adenoviruses. In animals, airborne disease transmission poses numerous threats to their protection, with major fatalities. The use of volumetric DBD was found to be effective in the inactivation of airborne porcine reproductive and respiratory syndrome viruses in a wind tunnel within milliseconds. Importantly, virus inactivation is boosted by short-term species during *in situ* droplet treatment [111].

After respiratory viruses, enteric viruses have the highest rate of infectivity and transmission. The virus decontamination process using cold plasma is a potential tool for food decontamination in the food industry. A DBD plasma torch using air gas was successfully used in a recent study to inactivate feline calicivirus (FCV), which is a surrogate of foodborne human norovirus [112]. The authors of the study also showed plasma-generated ROS as the key cause of viral inactivation. Another group of researchers assessed the antiviral activity of radio-frequency atmospheric plasma jets against FCV *in vitro*. Interestingly, they claimed that the decrease in virus titers was enhanced by an increase in exposure time and a reduced exposure gap. Similar to previous studies, the oxidized viral proteins (capsid) through plasma-generated RONS inside the solution were believed to be responsible for the observed anti-viral effect. This study highlights the importance of plasma efficacy in combating viral contamination in food [113]. Hepatitis A virus (HAV) and murine norovirus (MNV-1) in common fresh meats were inactivated within 10–20 min of plasma jet treatment. These findings revealed that 5 min of plasma jet exposure resulted in ~85–90% (1 $\log_{10}$ PFU/mL) and ~98–99% reduction (2 $\log_{10}$ PFU/mL) in HAV and MNV-1 titers, respectively, without affecting meat quality. This strategy can be utilized in raw meat handling and delivery processes to further protect fresh meat [114]. Another possible application of cold plasma in the food industry is the decontamination of food-handling surfaces and packaging materials. Over the past few years, the use of cold plasma has been widely accepted for decontamination, particularly for heat-labile surfaces and tissues, where chemical sanitization is not suitable. Nevertheless, few have reported on the inactivation of viruses, instead focusing on fungi and bacteria. Recently, the efficiency of cold plasma in decontaminating MNV-1 and HAV viruses on stainless steel surfaces has been shown [115]. Cumulative evidence and scavenger-based studies further reinforce the significant contribution of ROS in plasma-based antiviral activities.

Although most studies have focused on evaluating the potential of cold plasma for the inactivation of human respiratory viruses, some authors have focused on its efficacy in sexually transmitted viral diseases. Volotskova *et al* [116] showed that the pretreatment of macrophages with plasma blocks HIV virus-host cell fusion and integration. Remarkably, viral particles produced by plasma-treated cells had low infectivity, suggesting that the inhibitory effects of plasma was prolonged to the next phase of viral infection [116]. Similarly, another study showed the ability of the plasma jet to increase voltage and time of exposure thereby preventing the replication of HIV virions when helium feeding gas was used [117]. These studies rationalized the use of cold plasma as an anti-human immunodeficiency virus (HIV) treatment. Further studies have revealed that one of the human hepatitis B viruses (HBV) could be transmitted by contaminated apparatuses, which is considered a primary source of nosocomial infections. As it is difficult to cultivate in the laboratory, HBV decontamination is challenging to execute; thus, Vickery *et al* [118] used commercially available hydrogen peroxide gas plasma sterilizer on experimental surfaces contaminated with duck hepatitis B virus (DHBV) to mimic the likely transmission of contamination in sequential patients to inactivate DHBV. Following treatment with this plasma,

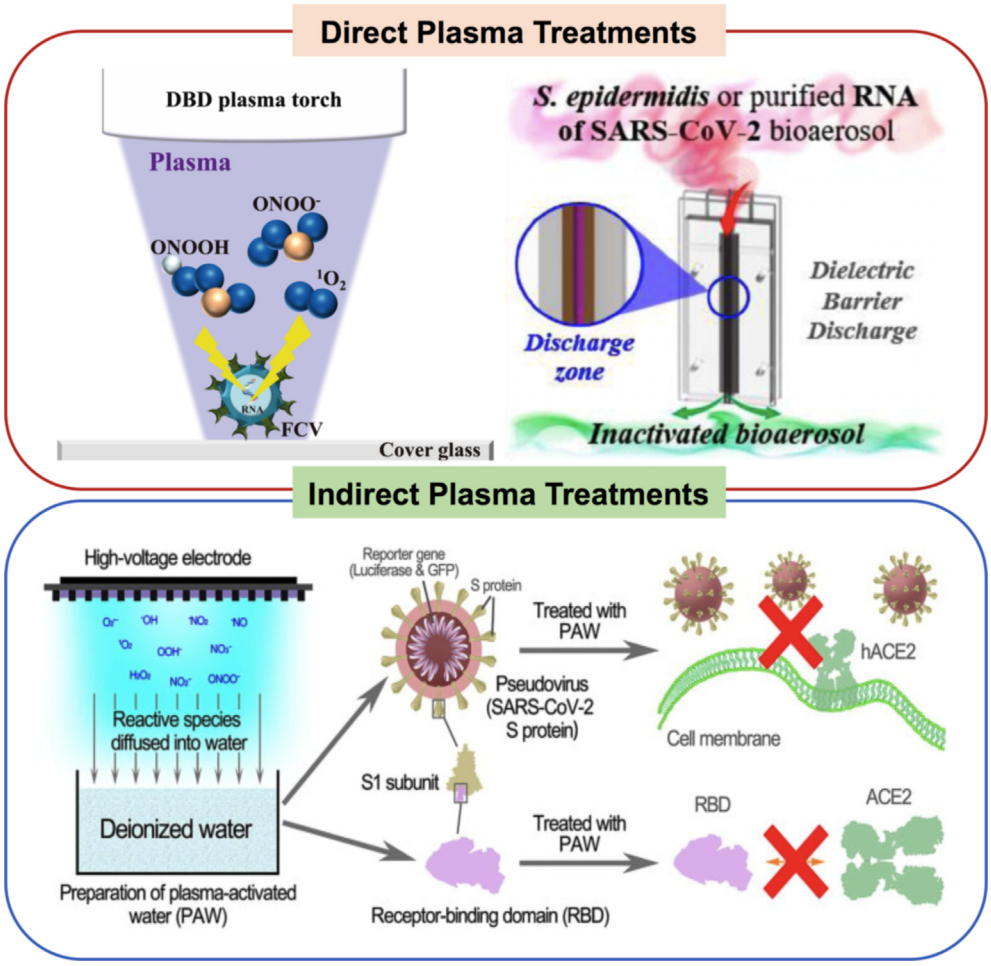

Fig. 4. Graphic illustration for the FCV-inactivation mechanism through DBD plasma torch [112]. Inactivation of aerosolized microdroplets for airborne indoor transmission by non-thermal plasma [129] PAW: an alternate decontaminator to destroy spike protein activity to inhibit SARS-CoV-2 [128].

sterilization of the instruments after a water wash was more than 60%.

Plasma technique against corona viruses

Monitoring the current progress in this area of plasma-induced virus inactivation is crucial during the ongoing pandemic, which highlights the demand for alternate virus inactivation techniques or the modernization of current practices. In this section, we mainly discuss the safety and technical approaches associated with the use of cold plasma for corona virus inactivation. Notably, the possibility of infection increases with viral exposure, and the early viral load impacts the severity of the disease [119 120]. The infectivity rate of SARS-CoV2 is high because it replicates over all throat and mouth areas [121]. As an efficient drug treatment for SARS-CoV2 infection is not available, the parochial treatment of the infected mucosa should be considered. Therefore, the use of advanced plasma technology is a promising option. Numerous reports have suggested that the use of plasma treatment in air is beneficial for pollution control, resulting in a decrease in airborne pathogens as mentioned above [122 123 124]. Furthermore, plasma-treated gas is effective for the contagious decontamination of various surfaces [125 126]. Based on these results, plasma technology has been applied to reduce or eradicate viral loads in the oral cavity of ventilated patients. The use of plasma-induced oxidation of cysteine is also recommended for the modification of SARS-CoV2 pathogenicity, and could potentially be delivered even via anesthetic masks, possibly at the time of surgery. However, mucosal tissue compatibility must be verified beforehand to prevent critical local side effects and tissue impairment.

In addition to direct plasma treatment, PAS can be used in the disinfection of surgical devices (Fig. 4). Complete inactivation efficiency of Newcastle disease virus by PAS, such as 0.9% NaCl, water, and 0.3% H_2O_2 after 30 min of treatment has been observed. Reactive oxygen nitrogen species were found to be the main cause of viral RNA degradation in smaller fragments. This study highlights the importance of PAS as a green disinfection invention and an alternative tool for sterilization in hospitals and public places, as well as stock farming applications where conventional chemical chlorine-based sanitizers, which are associated with risks of carcinogenic product formation with environmental pollutants, are used. These findings provide a basis for the function of PAS in addressing public health issues and eco-friendly sanitation [127]. Plasma activated solutions have also been used to understand its inactivation efficiency and mechanisms of the SARS-CoV2 virus. This study also concluded that the presence of reactive species was the main cause of viral cell death. They found that ROS of PAS damaged the RBD domain, which is positioned at the topmost margin of the spike protein and is also a very important part of the viral protein structure in SARS-CoV2 [128].

Previous studies have shown that plasma treatment of nitrogen and oxygen in the air results in the production of nitrogen oxides and O_3. Both these species are needed for viral inactivation on one part; nonetheless, they are highly toxic to the lungs at high concentrations. Consequently, it could be difficult to equalize the

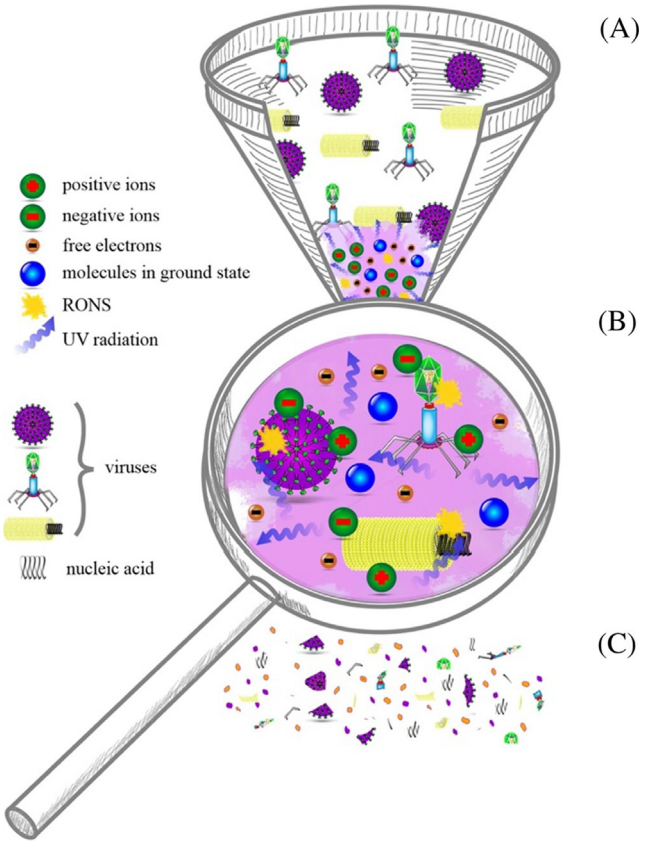

Fig. 5. Schematic representation of virus inactivation mechanism by plasma. (A) Morphological distinguishing viruses exposed with plasma. (B) Zoom of plasma characteristics accountable for virus inactivation. (C) Following plasma exposure, these virus particles are moderately or entirely damaged to become non-infective elements [107].

concentrations of these species to achieve appropriate antiviral effectiveness by minimizing such side effects. Additionally, other topics frequently under conversation are functions of plasma during hand disinfection. It is not unexpected that this approach could be applied to minimize SARS-CoV2 infections. Similar to established skin tolerability, the cold plasma potentiality with regard to the robust requirements for skin and hand methods must be validated [130]. When operating plasmas for virus inactivation, it is essential to fix the acceptable considerations and decide the exposure intervals that permit elements to cooperate with the spoiled material or surfaces. Various plasma sources can inactivate or considerably lower the infection rates of different pathogenic viruses under diverse conditions (Fig. 5). However, virus inactivation is extremely reliant on handling characteristics, and ideal factors must be preferred on a case-by-case basis [107].

Conclusion and future prospects

The present spread of viral disease has caused major public concern; to combat this, we must find a successful method for virus inactivation. Scientists worldwide are actively working to formulate various vaccines to immunize humans against different viruses. However, to effectively lower the death rate, we also need to consider precautionary channels that reduce viral spread. Apart from other physical techniques, plasma can be utilized as an eco-friendly treatment for the inactivation of different viruses in assorted matrices. During plasma application, RONS are responsible for viral inactivation by disrupting nucleic acids and capsid proteins (Fig. 6). Specifically, possible plasma application as an antiviral agent could significantly affect recent viral outbreaks (such as SARS-CoV-2) and could aid in alleviating the sanitization problems caused by forthcoming outbreaks.

However, more precise methods and further investigations will offer insight on which plasma elements are fundamental in every exposure and in what way they accurately disrupt viruses. We predict that plasma will be a promising candidate for virus inactivation in the near future. Eventually, the use of plasma could result in lowering human infection rates and drop-in cost-effective loads. In every circumstance, it is essential to estimate the possible unfavorable genotoxic consequences of plasma or PAS on humans. Although preliminary outcomes are favorable, the use of cold

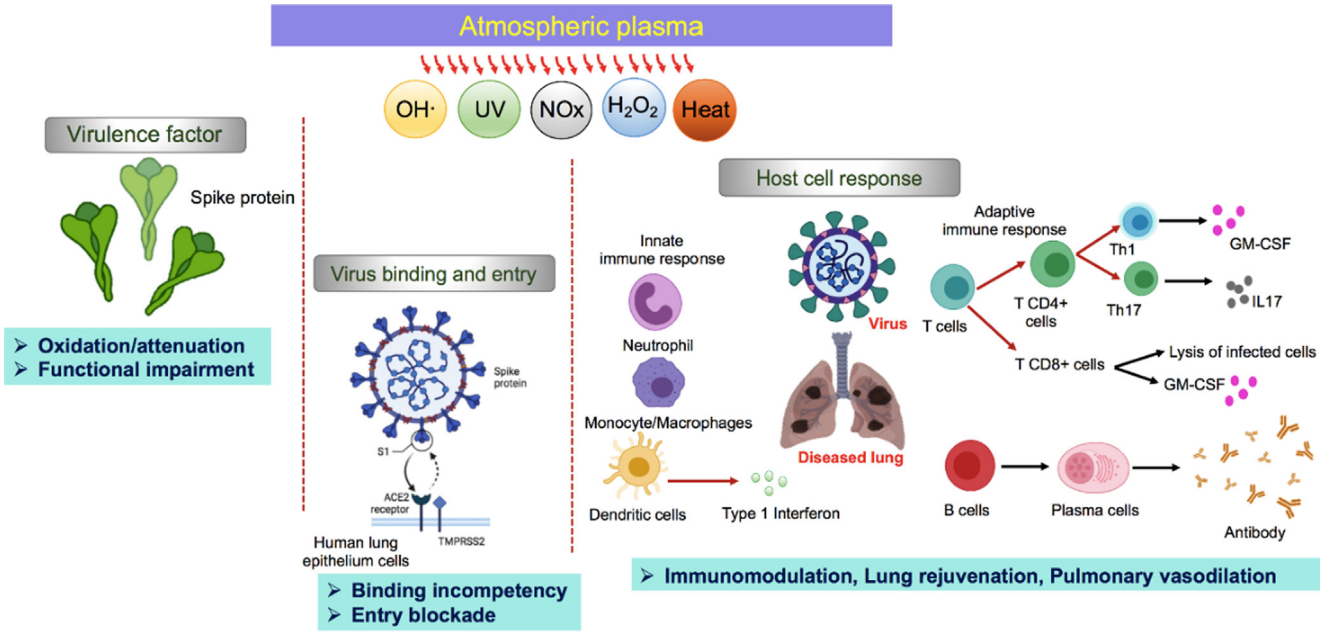

Fig. 6. Targets for cold plasma-based virus inactivation (virulence factor attenuation) and immunomodulation.

plasma in therapies or vaccine formulations still require substantial investigation prior to implementation.

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Compliance with Ethics Requirements

This article does not contain any studies with human or animal subjects.

CRediT authorship contribution statement

Neha Kaushik: Conceptualization, Software, Writing – original draft. **Sarmistha Mitra:** Writing – original draft. **Eun Jung Baek:** Writing – review & editing. **Linh Nhat Nguyen:** Writing – original draft. **Pradeep Bhartiya:** Writing – original draft, **June Hyun Kim:** Writing – review & editing. **Eun Ha Choi:** Writing – review & editing. **Nagendra Kumar Kaushik:** Conceptualization, Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

[1] Durmuş S, Ülgen KÖ. Comparative interactomics for virus-human protein-protein interactions: DNA viruses versus RNA viruses. FEBS Open Bio 2017;7(1):96–107. doi: https://doi.org/10.1002/2211-5463.12167.
[2] Davenport R, Schwarz L, Boulton J. The decline of adult smallpox in eighteenth-century London. Econ Hist Rev 2011;64:1289–314. doi: https://doi.org/10.1111/j.1468-0289.2011.00599.x.
[3] Frierson JG. The yellow fever vaccine: a history. Yale J Biol Med 2010;83:77–85.
[4] Taubenberger JK. The origin and virulence of the 1918 "Spanish" influenza virus. Proc Am Philos Soc 2006;150:86–112.
[5] Kaner J, Schaack S. Understanding Ebola: the 2014 epidemic. Global Health 2016;12:53. doi: https://doi.org/10.1186/s12992-016-0194-4.
[6] Wojda ThomasR, Valenza PamelaL, Cornejo K, McGinley T, Galwankar SagarC, Kelkar D, et al. The Ebola outbreak of 2014–2015: from coordinated multilateral action to effective disease containment, vaccine development, and beyond. J Glob Infect Dis 2015;7(4):127. doi: https://doi.org/10.4103/0974-777X.170495.
[7] Li H, Liu S-M, Yu X-H, Tang S-L, Tang C-K. Coronavirus disease 2019 (COVID-19): current status and future perspectives. Int J Antimicrob Agents 2020;55:105951. doi: https://doi.org/10.1016/j.ijantimicag.2020.105951.
[8] Hiscott J, Alexandridi M, Muscolini M, Tassone E, Palermo E, Soultsioti M, et al. The global impact of the coronavirus pandemic. Cytokine Growth Factor Rev 2020;53:1–9. doi: https://doi.org/10.1016/j.cytogfr.2020.05.010.
[9] Lee D, Moy N, Tritter J, Paolucci F. The COVID-19 pandemic: Global health policy and technology responses in the making. Health Policy Technol 2020;9(4):397–8. doi: https://doi.org/10.1016/j.hlpt.2020.10.001.
[10] Maarouf M, Rai KR, Goraya MU, Chen J-L. Immune ecosystem of virus-infected host tissues. Int J Mol Sci 2018;19:1379. doi: https://doi.org/10.3390/ijms19051379.
[11] Tan S-L, Ganji G, Paeper B, Proll S, Katze MG. Systems biology and the host response to viral infection. Nat Biotechnol 2007;25(12):1383–9. doi: https://doi.org/10.1038/nbt1207-1383.
[12] Chen X, Liu S, Goraya MU, Maarouf M, Huang S, Chen J-L. Host immune response to influenza A virus infection. Front Immunol 2018;9:320. doi: https://doi.org/10.3389/fimmu.2018.00320.
[13] Ciapponi A, Bardach A, Rey Ares L, Glujovsky D, Cafferata ML, Cesaroni S, Bhatti A. Sequential inactivated (IPV) and live oral (OPV) poliovirus vaccines for preventing poliomyelitis. Cochrane Database Syst Revi 2019;12:Cd011260. doi: https://doi.org/10.1002/14651858.CD011260.pub2.
[14] Preston R. The demon in the freezer. Fawcett Books; 2003.
[15] Ogholikhan S, Schwarz K. Hepatitis vaccines. Vaccines (Basel) 2016;4(1):6. doi: https://doi.org/10.3390/vaccines4010006.
[16] Afrough B, Dowall S, Hewson R. Emerging viruses and current strategies for vaccine intervention. Clin Exp Immunol 2019;196:157–66. doi: https://doi.org/10.1111/cei.13295.
[17] Graham BS. Advances in antiviral vaccine development. Immunol Rev 2013;255(1):230–42. doi: https://doi.org/10.1111/imr.12098.
[18] Lin Q, Lim JYC, Xue K, Yew PYM, Owh C, Chee PL, et al. Sanitizing agents for virus inactivation and disinfection. View 2020;1(2). doi: https://doi.org/10.1002/viw2.16.
[19] Li R, Cui L, Chen M, Huang Yu. Nanomaterials for airborne virus inactivation: a short review. Aerosol Sci Eng 2021;5(1):1–11.
[20] de Sanctis V, Canatan D, Corrons JLV, Karimi M, Daar S, Kattamis C, et al. Preliminary data on COVID-19 in patients with hemoglobinopathies: a multicentre ICET-A study. Mediterr J Hematol Infect Dis 2020;12:. doi: https://doi.org/10.4084/MJHID.2020.046e2020046.
[21] Peterhans E. Reactive oxygen species and nitric oxide in viral diseases. Biol Trace Elem Res 1997;56(1):107–16.
[22] Sies H, Berndt C, Jones DP. Oxidative stress. Annu Rev Biochem 2017;86(1):715–48. doi: https://doi.org/10.1146/annurev-biochem-061516-045037.
[23] Nikiforov A, Xiong Q, Britun N, Snyders R, Lu XP, Leys C. Absolute concentration of OH radicals in atmospheric pressure glow discharges with a liquid electrode measured by laser-induced fluorescence spectroscopy. Appl Phys Express 2011;4(2):026102. doi: https://doi.org/10.1143/APEX.4.026102.
[24] Kim YH, Hong YJ, Baik KY, Kwon GC, Choi JJ, Cho GS, et al. Measurement of reactive hydroxyl radical species inside the biosolutions during non-thermal atmospheric pressure plasma jet bombardment onto the solution. Plasma Chem Plasma Process 2014;34(3):457–72. doi: https://doi.org/10.1007/s11090-014-9538-0.
[25] Attri P, Kim YH, Park DH, Park JH, Hong YJ, Uhm HS, et al. Generation mechanism of hydroxyl radical species and its lifetime prediction during the plasma-initiated ultraviolet (UV) photolysis. Sci Rep 2015;5(1). doi: https://doi.org/10.1038/srep09332.
[26] Schmidt-Bleker A, Winter J, Iseni S, Dünnbier M, Weltmann K-D, Reuter S. Reactive species output of a plasma jet with a shielding gas device—combination of FTIR absorption spectroscopy and gas phase modelling. J Phys D Appl Phys 2014;47(14):145201. doi: https://doi.org/10.1088/0022-3727/47/14/145201.
[27] Gaens WV, Bogaerts A. Kinetic modelling for an atmospheric pressure argon plasma jet in humid air. J Phys D Appl Phys 2013;46(27):275201. doi: https://doi.org/10.1088/0022-3727/46/27/275201.
[28] Pei X, Lu Y, Wu S, Xiong Q, Lu X. A study on the temporally and spatially resolved OH radical distribution of a room-temperature atmospheric-pressure plasma jet by laser-induced fluorescence imaging. Plasma Sources Sci Technol 2013;22(2):025023. doi: https://doi.org/10.1088/0963-0252/22/2/025023.
[29] Mariotti D, Shimizu Y, Sasaki T, Koshizaki N. Gas temperature and electron temperature measurements by emission spectroscopy for an atmospheric microplasma. J Appl Phys 2007;101(1):013307. doi: https://doi.org/10.1063/1.2409318.
[30] Dupuis M, Fitzgerald G, Hammond B, Lester WA, Schaefer HF. Theoretical study of the H+O3↔OH+O2↔O+HO2 system. J Chem Phys 1986;84(5):2691–7. doi: https://doi.org/10.1063/1.450342.
[31] Bienert GP, Møller ALB, Kristiansen KA, Schulz A, Møller IM, Schjoerring JK, et al. Specific aquaporins facilitate the diffusion of hydrogen peroxide across membranes. J Biol Chem 2007;282(2):1183–92. doi: https://doi.org/10.1074/jbc.M603761200.
[32] Yan Jiupiang, Tsuichihara Nobue, Etoh Takeomi, Iwai Sumio. Reactive oxygen species and nitric oxide are involved in ABA inhibition of stomatal opening. Plant, Cell Environ 2007;30(10):1320–5. doi: https://doi.org/10.1111/j.1365-3040.2007.01711.x.
[33] Brooks AL, Hoel DG, Preston RJ. The role of dose rate in radiation cancer risk: evaluating the effect of dose rate at the molecular, cellular and tissue levels using key events in critical pathways following exposure to low LET radiation. Int J Radiat Biol 2016;92(8):405–26. doi: https://doi.org/10.1080/09553002.2016.1186301.
[34] Bhardwaj SK, Singh H, Deep A, Khatri M, Bhaumik J, Kim K-H, et al. UVC-based photoinactivation as an efficient tool to control the transmission of coronaviruses. Sci Total Environ 2021;792:148548. doi: https://doi.org/10.1016/j.scitotenv.2021.148548.
[35] Lytle CD, Sagripanti J-L. Predicted inactivation of viruses of relevance to biodefense by solar radiation. J Virol 2005;79(22):14244–52. doi: https://doi.org/10.1128/JVI.79.22.14244-14252.2005.
[36] Rastogi RP, Richa, Kumar A, Tyagi MB, Sinha RP. Molecular mechanisms of ultraviolet radiation-induced DNA damage and repair. J Nucleic Acids 2010;2010:1–32.
[37] Rauth AM. The physical state of viral nucleic acid and the sensitivity of viruses to ultraviolet light. Biophys J 1965;5(3):257–73.
[38] Lytle CD, Aaronson SA, Harvey E. Host-cell reactivation in mammalian cells. II. Survival of herpes simplex virus and vaccinia virus in normal human and xeroderma pigmentosum cells. Int J Radiat Biol Relat Stud Phys Chem Med 1972;22:159–65.
[39] Naunovic Z, Lim S, Blatchley ER. Investigation of microbial inactivation efficiency of a UV disinfection system employing an excimer lamp. Water Res 2008;42(19):4838–46. doi: https://doi.org/10.1016/j.watres.2008.09.001.
[40] Sarkis-Onofre R, Borges RdC, Demarco G, Dotto L, Schwendicke F, Demarco FF. Decontamination of N95 respirators against SARS-CoV-2: A scoping review. J Dent 2021;104:103534. doi: https://doi.org/10.1016/j.jdent.2020.103534.

[41] Sullivan R, Fassolitis AC, Larkin EP, Read Jr RB, Peeler JT. Inactivation of thirty viruses by gamma radiation. Appl Microbiol 1971;22(1):61–5.
[42] Hume A, Ames J, Rennick L, Duprex W, Marzi A, Tonkiss J, et al. Inactivation of RNA viruses by gamma irradiation: a study on mitigating factors. Viruses 2016;8(7):204. doi: https://doi.org/10.3390/v8070204.
[43] Donya M, Radford M, ElGuindy A, Firmin D, Yacoub MH. Radiation in medicine: origins, risks and aspirations. Glob Cardiol Sci Pract 2014;2014:437–48. doi: https://doi.org/10.5339/gcsp.2014.57.
[44] Feldmann F, Shupert WL, Haddock E, Twardoski B, Feldmann H. Gamma irradiation as an effective method for inactivation of emerging viral pathogens. Am J Trop Med Hyg 2019;100:1275–7. doi: https://doi.org/10.4269/ajtmh.18-0937.
[45] Kuefner M, Brand M, Engert C, Schwab S, Uder M. Radiation induced DNA double-strand breaks in radiology. In: Proceedings of RöFo-Fortschritte auf dem Gebiet der Röntgenstrahlen und der bildgebenden Verfahren; pp. 872–78. .
[46] Libertin CR, Panozzo J, Groh KR, Chang-Liu C-M, Schreck S, Woloschak GE. Effects of gamma rays, ultraviolet radiation, sunlight, microwaves and electromagnetic fields on gene expression mediated by human immunodeficiency virus promoter. Radiat Res 1994;140:91–6.
[47] Farkas J. Irradiation as a method for decontaminating food: a review. Int J Food Microbiol 1998;44(3):189–204.
[48] Brinston R, Wilson B. Converting to gamma-radiation sterilization: an overview for medical device manufacturers. Med Dev Technol 1993;4:18–22.
[49] Feng K, Divers E, Ma Y, Li J. Inactivation of a human norovirus surrogate, human norovirus virus-like particles, and vesicular stomatitis virus by gamma irradiation. Appl Environ Microbiol 2011;77:3507–17.
[50] Jordan RT, Kempe LL. Inactivation of some animal viruses with gamma radiation from cobalt-60. Proc Soc Exp Biol Med 1956;91(2):212–5. doi: https://doi.org/10.3181/00379727-91-22215.
[51] Latarjet R, Cramer R, Montagnier L. Inactivation, by UV-, X-, and γ-radiations, of the infecting and transforming capacities of polyoma virus. Virology 1967;33:104–11.
[52] Bryan WR, Lorenz E, Moloney JB. Studies on the effects in vitro of Roentgen radiation on the biological activity of the agent of chicken tumor I (Rous sarcoma). J Natl Cancer Inst 1950;10:1215–37.
[53] Decker C, Guir J, Kirn A. Infectivity and capacity for DNA replication of vaccinia virus irradiated by γ-rays. J Gen Virol 1969;4:221–7.
[54] Vilj M, Gottstein, Grozdani IC, Matanovj K, Miljani S, Mazija H, et al. Effect of low dose gamma-radiation upon Newcastle disease virus antibody level in chicken. Iran J radiat Res 2009:7:27–31..
[55] Sabbaghi A, Zargar M, Ghaemi A, Motamedi-Sedeh F, Zolfaghari MR. Inactivation of human influenza virus using gamma irradiation. J Arak Uni Med Sci 2019;22:112–23. doi: https://doi.org/10.32598/JAMS.22.4.100.
[56] Polley JR. Preparation of non-infective soluble antigens with gamma radiation. Can J Microbiol 1961;7(2):135–9.
[57] Adusumilli PS, Stiles BM, Chan M-K, Chou T-C, Wong RJ, Rusch VW, et al. Radiation therapy potentiates effective oncolytic viral therapy in the treatment of lung cancer. Ann Thoracic Surg 2005;80(2):409–17.
[58] Bidawid S, Farber JM, Sattar SA. Inactivation of hepatitis A virus (HAV) in fruits and vegetables by gamma irradiation. Int J Food Microbiol 2000;57(1-2):91–7. doi: https://doi.org/10.1016/S0168-1605(00)00235-X.
[59] Abolaban FA, Djouider FM. Gamma irradiation-mediated inactivation of enveloped viruses with conservation of genome integrity: Potential application for SARS-CoV-2 inactivated vaccine development. Open Life Sci 2021;16:558–70.
[60] Feldmann F, Shupert WL, Haddock E, Twardoski B, Feldmann H. Gamma irradiation as an effective method for inactivation of emerging viral pathogens. Am J Trop Med Hyg 2019;100(5):1275–7.
[61] Haddock E, Feldmann F, Shupert WL, Feldmann H. Inactivation of SARS-CoV-2 laboratory specimens. Am J Trop Med Hyg 2021;104:2195–8. doi: https://doi.org/10.4269/ajtmh.21-0229.
[62] Jinia AJ, Sunbul NB, Meert CA, Miller CA, Clarke SD, Kearfott KJ, et al. Review of sterilization techniques for medical and personal protective equipment contaminated with SARS-CoV-2. IEEE Access 2020;8:111347–54. doi: https://doi.org/10.1109/ACCESS.2020.3002886.
[63] Wainwright M. Local treatment of viral disease using photodynamic therapy. Int J Antimicrob Agents 2003;21(6):510–20.
[64] Maisch T. Resistance in antimicrobial photodynamic inactivation of bacteria. Photochem Photobiol Sci: Off J Photochem Assoc Eur Soc Photobiol 2015;14(8):1518–26.
[65] Costa L, Faustino MA, Neves MG, Cunha A, Almeida A. Photodynamic inactivation of mammalian viruses and bacteriophages. Viruses 2012;4:1034–74. doi: https://doi.org/10.3390/v4071034.
[66] Castano AP, Demidova TN, Hamblin MR. Mechanisms in photodynamic therapy: part one—photosensitizers, photochemistry and cellular localization. Photodiagn Photodyn Ther 2004;1(4):279–93. doi: https://doi.org/10.1016/S1572-1000(05)00007-4.
[67] Li W-Y, Xu J-G, He X-W. Characterization of the binding of methylene blue to DNA by spectroscopic methods. Anal Lett 2000;33(12):2453–64. doi: https://doi.org/10.1080/00032710008543201.
[68] Baptista MS, Cadet J, Di Mascio P, Ghogare AA, Greer A, Hamblin MR, et al. Type I and type II photosensitized oxidation reactions: guidelines and mechanistic pathways. Photochem Photobiol 2017;93(4):912–9. doi: https://doi.org/10.1111/php.12716.
[69] Wiehe A, O'Brien JM, Senge MO. Trends and targets in antiviral phototherapy. Photochem Photobiol Sci 2019;18(11):2565–612. doi: https://doi.org/10.1039/C9PP00211A.
[70] Tauxe RV. Food safety and irradiation: protecting the public from foodborne infections. Emerg Infect Dis 2001;7:516.
[71] Afrough B, Eakins J, Durley-White S, Dowall S, Findlay-Wilson S, Graham V, et al. X-ray inactivation of RNA viruses without loss of biological characteristics. Sci Rep 2020;10(1):21431. doi: https://doi.org/10.1038/s41598-020-77972-5.
[72] Kroeger AV, Kempf JE. Inactivation of the influenza virus by low voltage roentgen rays. J Bacteriol 1959;77:237–46.
[73] Friedewald WF, Anderson RS. The effects of roentgen rays on cell-virus associations: findings with virus-induced rabbit Papillomas and fibromas. J Exp Med 1943;78:285–304. doi: https://doi.org/10.1084/jem.78.4.285.
[74] Simmons S, Carrion R, Alfson K, Staples H, Jinadatha C, Jarvis W, Sampathkumar P, Chemaly RF, Khawaja F, Povroznik M, et al. Disinfection effect of pulsed xenon ultraviolet irradiation on SARS-CoV-2 and implications for environmental risk of COVID-19 transmission. MedRxiv 2020. doi: https://doi.org/10.1101/2020.05.06.20093658.
[75] Pendyala B, Patras A, D'Souza D. Genomic modeling as an approach to identify surrogates for use in experimental validation of SARS-CoV-2 and HuNoVs inactivation by UV-C treatment. bioRxiv 2020. doi: https://doi.org/10.1101/2020.06.14.151290.
[76] Dong J-H-X-P. Stability of SARS coronavirus in human specimens and environment and its sensitivity to heating and UV irra-diation. Biomed Environ Sci 2003;16:246–55.
[77] Sabino CP, Sellera FP, Sales-Medina DF, Machado RRG, Durigon EL, Freitas-Junior LH, et al. UV-C (254 nm) lethal doses for SARS-CoV-2. Photodiagn Photodyn Ther 2020;32:101995.
[78] Inagaki H, Saito A, Sugiyama H, Okabayashi T, Fujimoto S. Rapid inactivation of SARS-CoV-2 with deep-UV LED irradiation. Emerging Microbes Infect 2020;9:1744–7.
[79] Simmons SE, Carrion R, Alfson KJ, Staples HM, Jinadatha C, Jarvis WR, et al. Deactivation of SARS-CoV-2 with pulsed-xenon ultraviolet light: Implications for environmental COVID-19 control. Infect Control Hosp Epidemiol 2021;42:127–30.
[80] Buonanno M, Welch D, Shuryak I, Brenner DJ. Far-UVC light (222 nm) efficiently and safely inactivates airborne human coronaviruses. Sci Rep 2020;10:10285.
[81] Welch D, Buonanno M, Grilj V, Shuryak I, Crickmore C, Bigelow AW, et al. Far-UVC light: A new tool to control the spread of airborne-mediated microbial diseases. Sci Rep 2018;8:2752.
[82] Kitagawa H, Nomura T, Nazmul T, Omori K, Shigemoto N, Sakaguchi T, et al. Effectiveness of 222-nm ultraviolet light on disinfecting SARS-CoV-2 surface contamination. Am J Infect Control 2021;49:299–301.
[83] Eickmann M, Gravemann U, Handke W, Tolksdorf F, Reichenberg S, Müller TH, et al. Inactivation of three emerging viruses–severe acute respiratory syndrome coronavirus, Crimean-Congo haemorrhagic fever virus and Nipah virus–in platelet concentrates by ultraviolet C light and in plasma by methylene blue plus visible light. Vox Sang 2020;115:146–51.
[84] Lytle CD. Host-cell reactivation in mammalian cells. Int J Radiat Biol Related Stud Phys, Chem Med 1972;22(2):167–74.
[85] Bedell K, Buchaklian AH, Perlman S. Efficacy of an automated multiple emitter whole-room ultraviolet-C disinfection system against coronaviruses MHV and MERS-CoV. Infect Control Hospital Epidemiol 2016;37:598–9.
[86] Darnell ME, Subbarao K, Feinstone SM, Taylor DR. Inactivation of the coronavirus that induces severe acute respiratory syndrome, SARS-CoV. J Virol Methods 2004;121:85–91.
[87] de Roda Husman AM, Bijkerk P, Lodder W, Van Den Berg H, Pribil W, Cabaj A, et al. Calicivirus inactivation by nonionizing (253.7-nanometer-wavelength [UV]) and ionizing (gamma) radiation. Appl Environ Microbiol 2004;70:5089–93.
[88] Jordan RT, Kempe LL. Inactivation of some animal viruses with gamma radiation from cobalt-60. Proc Soc Exp Biol Med 1956;91:212–5.
[89] Thomas F, Davies A, Dulac G, Willis N, Papp-Vid G, Girard A. Gamma ray inactivation of some animal viruses. Canadian J Comp Med 1981;45:397.
[90] House C, House JA, Yedloutschnig RJ. Inactivation of viral agents in bovine serum by gamma irradiation. Can J Microbiol 1990;36:737–40.
[91] Leung A, Tran K, Audet J, Lavineway S, Bastien N, Krishnan J. In vitro Inactivation of SARS-CoV-2 using gamma radiation. Appl Biosaf 2020;25:157–60.
[92] Perlin M, Mao JCH, Otis ER, Shipkowitz NL, Duff RG. Photodynamic inactivation of influenza and herpes viruses by hematoporphyrin. Antiviral Res 1987;7(1):43–51. doi: https://doi.org/10.1016/0166-3542(87)90038-6.
[93] Käsermann F, Kempf C. Photodynamic inactivation of enveloped viruses by buckminsterfullerene. Antiviral Res 1997;34:65–70. doi: https://doi.org/10.1016/s0166-3542(96)01207-7.
[94] Afrough B, Eakins J, Durley-White S, Dowall S, Findlay-Wilson S, Graham V, et al. X-ray inactivation of RNA viruses without loss of biological characteristics. Sci Rep 2020;10:21431.
[95] Ryan HA, Neuber J, Song S, Beebe SJ, Jiang C. Effects of a non-thermal plasma needle device on HPV-16 positive cervical cancer cell viability in vitro. In: Proceedings of 2016 38th Annual International Conference of the IEEE Engineering in Medicine and Biology Society (EMBC), 16–20 Aug. 2016, 537–40..

[96] Brullé L, Vandamme M, Riès D, Martel E, Robert E, Lerondel S, et al. Effects of a non thermal plasma treatment alone or in combination with gemcitabine in a MIA PaCa2-luc orthotopic pancreatic carcinoma model. PLoS ONE 2012;7 (12):e52653. doi: https://doi.org/10.1371/journal.pone.0052653.
[97] Zhen X, Sun H-N, Liu REN, Choi HS, Lee D-S. Non-thermal plasma-activated medium induces apoptosis of Aspc1 cells through the ROS-dependent autophagy pathway. In Vivo 2020;34:143–53.
[98] Feil L, Koch A, Brucker S, Wallwiener D, Weiss M. Inhibition of cervical cancer cell growth by non-thermal atmospheric plasma application utilizing a thermal argon plasma source. Eur J Obstet Gynecol Reprod Biol 2019;234: e131. doi: https://doi.org/10.1016/j.ejogrb.2018.08.446.
[99] Malyavko A, Yan D, Wang Q, Klein AL, Patel KC, Sherman JH, et al. Cold atmospheric plasma cancer treatment, direct versus indirect approaches. Mater Adv 2020;1(6):1494–505.
[100] Sakudo A, Yagyu Y, Onodera T. Disinfection and sterilization using plasma technology: fundamentals and future perspectives for biological applications. Int J Mol Sci 2019;20(20):5216. doi: https://doi.org/10.3390/ijms20205216.
[101] Weiss M, Daeschlein G, Kramer A, Burchardt M, Brucker S, Wallwiener D, et al. Virucide properties of cold atmospheric plasma for future clinical applications. J Med Virol 2017;89(6):952–9. doi: https://doi.org/10.1002/jmv.24701.
[102] Guo Li, Xu R, Gou Lu, Liu Z, Zhao Y, Liu D, et al. Mechanism of virus inactivation by cold atmospheric-pressure plasma and plasma-activated water. Appl Environ Microbiol 2018;84(17). doi: https://doi.org/10.1128/AEM.00726-18.
[103] Yasuda H, Miura T, Kurita H, Takashima K, Mizuno A. Biological evaluation of DNA damage in bacteriophages inactivated by atmospheric pressure cold plasma. Plasma Processes Polym 2010;7:301–8. doi: https://doi.org/10.1002/ppap.200900088.
[104] Xia T, Kleinheksel A, Lee EM, Qiao Z, Wigginton KR, Clack HL. Inactivation of airborne viruses using a packed bed non-thermal plasma reactor. J Phys D Appl Phys 2019;52(25):255201. doi: https://doi.org/10.1088/1361-6463/ab1466.
[105] Wu Y, Liang Y, Wei K, Li W, Yao M, Zhang J, et al. MS2 virus inactivation by atmospheric-pressure cold plasma using different gas carriers and power levels. Appl Environ Microbiol 2015;81(3):996–1002.
[106] Tanaka Y, Yasuda H, Kurita H, Takashima K, Mizuno A. Analysis of the inactivation mechanism of bacteriophage $\varphi\hbox{X}174$ by atmospheric pressure discharge plasma. IEEE Trans Ind Appl 2014;50 (2):1397–401.
[107] Filipić A, Gutierrez-Aguirre I, Primc G, Mozetič M, Dobnik D. Cold plasma, a new hope in the field of virus inactivation. Trends Biotechnol 2020;38 (11):1278–91. doi: https://doi.org/10.1016/j.tibtech.2020.04.003.
[108] Sakudo A, Shimizu N, Imanishi Y, Ikuta K. N₂ gas plasma inactivates influenza virus by inducing changes in viral surface morphology, protein, and genomic RNA. Biomed Res Int 2013;2013:. doi: https://doi.org/10.1155/2013/694269694269.
[109] Sakudo A. N 8322 gas plasma inactivates influenza virus mediated by oxidative stress. Front Biosci 2014;E6(1):69–79.
[110] Bunz O, Mese K, Zhang W, Piwowarczyk A, Ehrhardt A. Effect of cold atmospheric plasma (CAP) on human adenoviruses is adenovirus type-dependent. PLoS ONE 2018;13(10):e0202352. doi: https://doi.org/10.1371/journal.pone.0202352.
[111] Nayak G, Andrews AJ, Marabella I, Aboubakr HA, Goyal SM, Olson BA, et al. Rapid inactivation of airborne porcine reproductive and respiratory syndrome virus using an atmospheric pressure air plasma. Plasma Processes Polym 2020;17(10):1900269. doi: https://doi.org/10.1002/ppap.v17.1010.1002/ppap.201900269.
[112] Yamashiro R, Misawa T, Sakudo A. Key role of singlet oxygen and peroxynitrite in viral RNA damage during virucidal effect of plasma torch on feline calicivirus. Sci Rep 2018;8:17947. doi: https://doi.org/10.1038/s41598-018-36779-1.
[113] Aboubakr HA, Williams P, Gangal U, Youssef MM, El-Sohaimy SAA, Bruggeman PJ, et al. Virucidal effect of cold atmospheric gaseous plasma on feline calicivirus, a surrogate for human norovirus. Appl Environ Microbiol 2015;81(11):3612–22.
[114] Bae S-C, Park SY, Choe W, Ha S-D. Inactivation of murine norovirus-1 and hepatitis A virus on fresh meats by atmospheric pressure plasma jets. Food Res Int 2015;76:342–7. doi: https://doi.org/10.1016/j.foodres.2015.06.039.
[115] Park SY, Ha S-D. Assessment of cold oxygen plasma technology for the inactivation of major foodborne viruses on stainless steel. J Food Eng 2018;223:42–5. doi: https://doi.org/10.1016/j.jfoodeng.2017.11.041.
[116] Volotskova O, Dubrovsky L, Keidar M, Bukrinsky M. Cold atmospheric plasma inhibits HIV-1 replication in macrophages by targeting both the virus and the cells. PLoS ONE 2016;11(10):e0165322. doi: https://doi.org/10.1371/journal.pone.0165322.
[117] Amiran MR, Sepahi AA, Zabiollahi R, Ghomi H, Momen SB, Aghasadeghi MR. In vitro Assessment of Antiviral Activity of Cold Atmospheric Pressure Plasma Jet against the Human Immunodeficiency Virus (HIV). JoMMID 2016;4:62–7.
[118] Vickery K, Deva AK, Zou J, Kumaradeva P, Cossart LB. Inactivation of duck hepatitis B virus by a hydrogen peroxide gas plasma sterilization system: laboratory and 'in use' testing. J Hospital Infect 1999;41:317–22. doi: https://doi.org/10.1053/jhin.1998.0516.
[119] Heinzerling A, Stuckey MJ, Scheuer T, Xu K, Perkins KM, Resseger H, et al. Transmission of COVID-19 to health care personnel during exposures to a hospitalized patient — Solano County, California, February 2020. MMWR Morb Mortal Wkly Rep 2020;69(15):472–6. doi: https://doi.org/10.15585/mmwr.mm6915e5.
[120] Raoult D, Zumla A, Locatelli F, Ippolito G, Kroemer G. Coronavirus infections: Epidemiological, clinical and immunological features and hypotheses. Cell Stress 2020;4(4):66–75. doi: https://doi.org/10.15698/cst2020.04.216.
[121] Wölfel R, Corman VM, Guggemos W, Seilmaier M, Zange S, Müller MA, et al. Virological assessment of hospitalized patients with COVID-2019. Nature 2020;581(7809):465–9. doi: https://doi.org/10.1038/s41586-020-2196-x.
[122] Karatum O, Deshusses MA. A comparative study of dilute VOCs treatment in a non-thermal plasma reactor. Chem Eng J 2016;294:308–15. doi: https://doi.org/10.1016/j.cej.2016.03.002.
[123] Müller S, Zahn R-J. Air pollution control by non-thermal plasma. Contrib Plasma Phys 2007;47(7):520–9. doi: https://doi.org/10.1002/ctpp.200710067.
[124] Brandenburg R, Kovačević VV, Schmidt M, Basner R, Kettlitz M, Sretenović GB, et al. Plasma-based pollutant degradation in gas streams: status, examples and outlook. Contrib Plasma Phys 2014;54(2):202–14. doi: https://doi.org/10.1002/ctpp.201310059.
[125] Bhatt S, Mehta P, Chen C, Schneider CL, White LN, Chen H-L, et al. Efficacy of low-temperature plasma-activated gas disinfection against biofilm on contaminated GI endoscope channels. Gastrointest Endosc 2019;89 (1):105–14. doi: https://doi.org/10.1016/j.gie.2018.08.009.
[126] Schnabel U, Schmidt C, Stachowiak J, Bösel A, Andrasch M, Ehlbeck J. Plasma processed air for biological decontamination of PET and fresh plant tissue. Plasma Processes Polym 2018;15(2):1600057.
[127] Su X, Tian Y, Zhou H, Li Y, Zhang Z, Jiang B, et al. Inactivation efficacy of nonthermal plasma-activated solutions against newcastle disease virus. Appl Environ Microbiol 2018;84(9):e02836–17. doi: https://doi.org/10.1128/AEM.02836-17.
[128] Guo Li, Yao Z, Yang Lu, Zhang H, Qi Yu, Gou Lu, et al. Plasma-activated water: An alternative disinfectant for S protein inactivation to prevent SARS-CoV-2 infection. Chem Eng J 2021;421:127742.
[129] Bisag A, Isabelli P, Laurita R, Bucci C, Capelli F, Dirani G, et al. Cold atmospheric plasma inactivation of aerosolized microdroplets containing bacteria and purified SARS-CoV-2 RNA to contrast airborne indoor transmission. Plasma Processes Polym 2020;17(10):2000154. doi: https://doi.org/10.1002/ppap.v17.1010.1002/ppap.202000154.
[130] Bekeschus S, Kramer A, Suffredini E, von Woedtke T, Colombo V. Gas plasma technology—an asset to healthcare during viral pandemics such as the COVID-19 crisis?. IEEE Trans Radiat Plasma Med Sci 2020;4(4):391–9.

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