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Review on Inactivation of Airborne Viruses using Non-Thermal Plasma technologies: From MS2 to Coronavirus

Imen ASSADI¹, Ahlem GUESMI², Oussama BAALOUJ³, Hichem ZEGHIOUD⁴,
Walid ELFALLEH¹, Naoufel BENHAMMADI², Lotfi KHEZAMI², Aymen ASSADI^{5,*}

¹Laboratoire Energie, Eau, Environnement et Procèdes, LR18ES35, ENIG, Université de Gabès, 6072
Gabès, Tunisia.

²Department of Chemistry, Imam Mohammad Ibn Saud Islamic University P.O. Box 5701, Riyadh, 11432,
Saudi Arabia.

³Laboratory of Reaction Engineering, Faculty of Mechanical Engineering and Process Engineering, USTHB,
BP 32, 16111 Algiers, Algeria.

⁴Department of Process Engineering, Badji Mokhtar University, P.O. Box 12, 23000, Annaba, Algeria

⁵Univ. Rennes ; ENSCR- 11 Allée de Beaulieu - Rennes 35708; France

*Corresponding Author: Phone: +33 (0) 2 23 23 81 52 (Office) Email: Aymen.Assadi@ensc-rennes.fr

Abstract

Although several non-thermal plasmas (NTPs) technologies have been widely investigated in air treatment, very few studies have focused on the inactivation mechanism of viruses by NTPs. Due to its efficiency and environmental compatibility, non-thermal plasma could be considered a promising virus-inactivation technology. Plasma is a partly or fully ionized gas including some species (i.e., electrons, free radicals, ions, and neutral molecules) to oxidize pollutants or inactivate harmful organisms. Non-thermal plasmas are made using less energy and have an active electron at a much higher temperature than bulk gas molecules. This review describes NTPs for virus inactivation in indoor air. The different application processes of plasma for microorganism inactivation at both laboratory and pilot-scale was also reviewed. This paper reports on recent advances in this exciting area of viral inactivation, identifying applications and mechanisms of inactivation, and summarizing the results of the latest experiments in the literature. Moreover, special attention was paid to the mechanism of virus inactivation. Finally, the paper suggests research directions in the field of Airborne Viruses inactivation using Non-Thermal Plasma.

Keywords: Non-thermal plasma, virus inactivation, indoor air, best-advanced oxidation.

31 1. Introduction

32 Viruses are part of the life history on our planet and the most abundant and diverse
33 microbes. These species dwelled in the World for billions of years and infected all cell-
34 based life forms organisms, i.e., from bacteria to humans, animals, and plants
35 (Couch,1981). Consequently, they are responsible for numerous hospitalizations, animal
36 diseases, deaths, and crop losses, leading to immense social, economic, and biological
37 pressures (Filipić et al., 2020a). They have adapted to various climates and are now found
38 in all environmental compartments, such as the air, where the risk of infectious respiratory
39 diseases is higher than any other sources (Tregoning and Schwarze, 2010). A viral
40 respiratory infection could be exposed to bacterial disease, thus duplicating the risk of
41 death (Hament et al., 1999). Despite considerable public health efforts, epidemics of viral
42 respiratory infections are still common (widespread) in healthy humans and can lead to the
43 death of vulnerable people (Moriyama et al. 2020). In 2017 alone, the Global Burden of
44 Disease study found that air pollution is a fatal source of nearly 5 million premature deaths
45 (Xiao Wu, Rachel C Nethery, M Benjamin Sabath, Danielle Braun, 2020). Also, another
46 study *via* the World Bank in 2007 found that air infection diseases kill 750 000 people
47 annually in China (Chang, 2012).

48 Typically, Viruses exist in the air at specific concentrations, which are not generally
49 sufficient to cause disease in humans because the immune system of healthy humans
50 prevents infections. However, the risk of human contamination rises significantly at higher
51 infectious virus concentrations (Nikitin et al., 2014). Numerous epidemiological studies
52 demonstrated that air transmissions via aerosols, large droplets, or direct contact with
53 secretions lead to a substantial increase in human morbidity and mortality, making air the
54 most significant vector of viral infection (Couch R et al., 1981),(Martínez-Montelongo et
55 al., 2020]. Pollution of indoor and outdoor air has been identified as responsible for
56 significant deleterious effects on human health and social and economic growth (Martínez-
57 Montelongo et al., 2020). However, indoor air pollution is the primary concern as it is 2-5
58 times more polluted than outside air pollution; in extreme cases, it can be over 100 times
59 higher (Fang et al., 2016). Also, indoor environments often harbor potentially unsafe
60 microorganisms (Giuseppina La Rosa, Marta Fratini, and Muscill, 2013).

61 . Pandemics such as the COVID-19 established the necessity for indoor air cleaning
62 and air purification techniques (Kim and Jang 2018) and replacing, completing, or updating
63 existing conventional viral inactivation methods (Li et al., 2020). Most decontamination

64 processes commonly used for viral inactivation like photocatalysis (TiO_2 +UVA) and
65 filtration don't have an effective disinfection ability against viruses and have substantial
66 inconveniences (Zhang and Gamage, 2010). Indeed, the most applied technologies to
67 control and remove airborne pathogens are the collection of bioaerosols on filters, such as
68 Porcine Reproductive and Respiratory Syndrome (PRRS), High-Efficiency Particulate
69 Arrestance (HEPA), Minimum Efficiency Reporting Value (MERV) 14 and MERV 16 filters,
70 UV germicidal irradiation (UVGI), Atmospheric Pressure Non-Thermal Plasmas (APNTP),
71 and Cold Atmospheric Pressure Plasma (CAPP). HEPA filtration has been experienced to
72 be very effective in removing bioaerosols from airstreams. In contrast, MERV filters were
73 exhibited to be the safer choice for both roof bolter and face drill (Cecala et al., 2016).

74 Indeed, Farnsworth et al. (Farnsworth et al., 2006) showed the highest efficiency (i.e.,
75 $96.5 \pm 1.5 \%$) of the bacterial collection of *B. subtilis* by HEPA filtration. Alonso et al. (Alonso
76 et al. 2013) reassessed the impact of air filtration from commercial sow farms on the
77 incidence of PRRSV introductions. They found that air filtration resulted in a reduction of
78 approximately 80 % of the risk with introducing the new PRRSv, thus highlighting the role
79 of PRRSv in aerosol filtration of the air. Otherwise, using HEPA filters denotes several
80 disadvantages, such as high filter replacement costs, and high-pressure drops and
81 increased energy consumption to maintain the desired ventilation rate.

82 Likewise, HEPA filters do not deactivate pathogens, hence the possibility of re-
83 emitting more resilient pathogens into the ambient air during the filter replacement and
84 disposal processes (Miaskiewicz-Peska and Lebkowska, 2012). Effective UVGI
85 disinfection requires enough high illumination intensity, achieved via an association of
86 several bulbs with suitable exposure times in a well-designed airing system. However, the
87 application of UVGI in the disinfection of indoor air is limited through the necessity to avoid
88 the harmful effects of UV on human and animal health (Bolashikov and Melikov 2009). One
89 of those drawbacks is contamination with by-products of disinfection with carcinogenic
90 effects and environmental pollution (Su et al., 2018). Unlike those chemical methods,
91 NTPs, also named cold plasma (CP), entered the decontamination processes as a new,
92 effective, clean solution for inactivating the virus (Lacombe et al., 2017). NTP is a complex
93 mixture, consisting of a partially ionized gas containing high-energy photons (UV), free
94 electrons, ions, radicals, and other reactive species (Assadi et al., 2015; Jia et al. 2015;
95 Schmidt and Jo 2015; Jiang et al., 2017; Schiappacasse et al., 2020). It produces high-
96 energy electrons which bump gas molecules, driving their dissociation, excitation, and

97 ionization (Huang et al., 2018), thereby occurring many reactive oxidative species (ROS)
98 including $\cdot\text{OH}$, O_2^- , O_3 , and H_2O_2 . This ROS may effectively inactivate viruses (Li et al.,
99 2020), as well as nitrogen species (NS) which play vital roles in viruses inactivation (Wang
100 et al., 2016). It is one of the advanced oxidation technologies proven effective in biomedical
101 fields, including decontamination and sterilization, due to its low cost, easy operation, and
102 wide range of applications (Feng et al. 2020). It is also essentially clean and generates
103 only small amounts of persistent chemical species (Moreau et al. 2008). Few papers report
104 NTP as an anti-viral treatment in the air, yet it shows great promise and has proven to
105 inactivate many types of microorganisms such as airborne viruses (Gallagher et al. 2004;
106 Scholtz et al., 2015). For instance, NTP inactivation of coronavirus (COVID-19) (Chen and
107 Wirz, 2020), Newcastle disease virus (NDV) (Su et al., 2018), and avian influenza virus
108 (AIV) (Wang et al., 2016) have been successful. Thus, strategies should continue to be
109 developed to improve NTP-based technologies to battle viruses for humanity's long-term
110 benefit.

111 This review focuses on NTPs for the inactivation of viruses in indoor air. It is an
112 attractive advanced oxidation technology since it is a new, effective, clean solution for
113 inactivating the virus and presents recent advances in the exciting area of virus's
114 inactivation, identifying applications and inactivation mechanisms, and summarizing
115 findings in the latest literature experiments. An enumeration of the worldwide problem
116 related to Airborne Viruses is cited. The effect of key operating parameters on NTP plasma
117 techniques used in the literature is developed.

118 **2. Viruses in indoor air**

119 The term "indoor" includes all enclosed spaces occupied by humans such as
120 homes, offices, schools, workplaces, commercial units, airports, hospitals, other health
121 facilities, and so on (Moreau et al., 2008). According to the Environmental Protection
122 Agency (EPA), many people spend about 90 % or more (i.e., most of their time indoors)
123 (Chen et al., 2020). People spend more time at home than in any other indoor environment
124 (Scholtz et al., 2015). Also, indoor air pollution is 2-5 times more polluted than outside air
125 pollution. In extreme cases, it can be over 100 times higher (Chen et al., 2020) because of
126 the various sources of indoor chemical pollution like formaldehyde from wood and biologic
127 pollution (Anderson et al., 1998). Besides, a study estimated the total concentrations of
128 viruses in indoor air and found about 10^5 particles m^{-3} , which is an extremely high

129 concentration (Chen et al., 2020). Environmental Protection Agency (EPA) ranks indoor air
130 pollution as one of the top 5 environmental hazards and health threats, improving indoor
131 air quality (IAQ) to reduce sickness. IAQ is the field of applied science or analysis of indoor
132 air conditions that includes the viral, microbiological, and chemical-physical composition in
133 the air in and around buildings (Maisey et al., 2012). The definition of acceptable IAQ is
134 provided by standard ISO 16000 "Air in confined environments" in particular ISO 16814:
135 air in an internal building which does not displease the vast majority of people inside to
136 ensure them healthy conditions and is unlikely to contain pollutants leading to a risk to
137 exposure to health (Prussin et al., 2015). The World Health Organization (WHO) stated
138 that IAQ is essential for preserving human health (Australian Building codes Board, 2018).

139 Indoor air quality has a significant impact on human health status (Australian
140 Building codes Board, 2018). Therefore, WHO concluded that it is the human right to
141 breathe healthy indoor air (Capolongo et al., 2017). It is, therefore, the responsibility of all
142 those involved to ensure acceptable indoor air quality (Abouleish, 2020). It becomes just
143 like an invisible killer (WHO, 2000). According to China Standardization Association, 68 %
144 of illnesses are caused by poor indoor air quality (Vohra et al., 2006). In addition, some
145 authors reported that higher disease and mortality rates are linked to air pollution and poor
146 indoor air quality [46]. Yet, exposure to air pollutants can increase the susceptibility and
147 severity of respiratory viruses in indoor air (Capolongo et al., 2017). Most upper respiratory
148 infections (URIs) like cold, pneumonia, pharyngitis, laryngitis, and epiglottitis are caused
149 by over 200 different viruses, including influenza viruses, rhinoviruses, coronaviruses,
150 adenoviruses, respiratory syncytial viruses, and enteroviruses (US EPA, 2006). Recently,
151 other viruses emerged at the end of 2019, such as SARS-CoV-2, killing millions of people
152 worldwide, thus demonstrating the dangerousness of infectious respiratory diseases
153 (Bourouiba, 2020). Due to their high airborne transmission ability, inhaling these viruses
154 can cause or worsen disease in humans (Rahmani et al., 2020), especially in crowded and
155 poorly ventilated environments. They are responsible for most upper respiratory infections
156 (Cimbala 2003). Many of the illnesses associated with SARS-CoV-2 that increase the risk
157 of death are those caused by long-term exposure to air pollution (Xiao Wu, Rachel C
158 Nethery, M Benjamin Sabath, Danielle Braun, 2020). Indoor virus species is one of the
159 most common environmental health problems reported today (Table1). Exposure to
160 aerosols generated by coughing and sneezing from infected individuals or in contact with
161 droplet-contaminated surfaces (plastic, metal, and clothing) have been widely viewed as

162 the dominant mode of transmission of respiratory pathogens (Virus and bacteria). In
 163 addition, it has been validated that airborne transmission is traditionally defined as
 164 involving the inhalation of infectious aerosols primarily at a distance of less than 1 to 2 m
 165 from the individual carrying the virus. Such transmission has been reported, and
 166 considered as the main route of virus transmission. However, recently airborne
 167 transmission of many respiratory viruses has been validated, such as coronavirus (SARS-
 168 CoV), Middle East respiratory syndrome (MERS) -CoV, influenza virus (Wang et al., 2021).

169 Table 1: Not exhaustive list on types of viruses and their effects on human health

Type of virus	Effects on human health	Ref
<i>SARS-CoV-2</i>	SARS-CoV-2 does not damage only the respiratory system and lungs; it can infect the urogenital system, nervous system, digestive system, and circulatory system. It can also cause an Inflammatory Cascade. Among its symptoms are mild disease, severe lung injury, and multi-organ failure leading to death, especially in older patients with other comorbidities. Other significant outcomes of coronavirus infection are acute respiratory distress syndrome and pulmonary fibrosis. The infection's long-term implications on human health are unknown, like the virus's possible impacts on cellular lifetime as well as organismal healthspan, which can cause diseases such as Parkinson's and other neurodegenerative illnesses.	(Day et al. 2020; Liang et al. 2020; Lippi et al. 2020; Monteil et al. 2020; Zhang et al. 2020)
<i>Rhinoviruses</i>	The main site of RV infection is the nasal mucosa. It is a common cause of colds (acute nasopharyngitis), which is a mild disease of the upper respiratory tract. It can also cause chronic obstructive pulmonary disease, asthma, or cystic fibrosis might become life-threatening.	(Myatt et al. 2004; Turner 2007; Ashraf et al. 2013; Blaas and Fuchs 2016; Thibaut et al. 2016; To Kelvin et al. 2017)
<i>MS2 Bacteriophage</i>	MS2 is an enteric virus; it is non-hazardous to humans and animals and is widely utilized as a surrogate for pathogenic viruses in aerosol investigations. It is an example of Noroviruses which are highly infectious and are associated with sporadic gastroenteritis.	(Olson et al. 2004; Dawson et al. 2005; Tung-Thompson et al. 2015)
<i>Influenza viruses</i>	Influenza is a highly contagious viral respiratory disease. A dry cough, headaches, muscular and joint discomfort, and general weariness are	(Hayden et al. 1998; Brankston et al. 2007; Carrat et

(types A, B, C)	among the symptoms. The flu can become dangerous, requiring hospitalization or even death.	al. 2008; World Health Organization 2019)
<i>Varicella viruses</i>	Varicella virus, one of the five human herpes viruses, causes two distinct diseases, varicella (chickenpox) and shingles (herpes zoster). Its's symptoms are fever concurrent with a self-limiting rash on the skin and sometimes mucosa. Headache, malaise, and loss of appetite.	(Mueller et al. 2008; Andrei and Snoeck 2013; Sauerbrei 2016)
<i>Measles virus</i>	Measles is a disease that can infect only humans; it attacks immunological cells, leading to a rapid deterioration of the immune system. It can cause three different types of encephalitis or central nervous system (CNS) diseases which are acute postinfectious encephalitis, acute progressive infectious encephalitis, and the lethal neurodegenerative disease subacute sclerosing panencephalitis.	(Bellini et al. 1994; Norrby and Kristensson 1997; Mrkic et al. 1998; Reuter and Schneider-Schaulies 2010; Mina et al. 2019)
<i>Hantavirus</i>	Hantavirus pulmonary syndrome is a zoonotic disease; its transmission to humans can lead to Hemorrhagic fever with renal syndrome or nephropathy epidemic named hantavirus pulmonary syndrome, also leading to disease with severe cardiopulmonary.	(Khaiboullina and St. Jeor 2002; Muranyi et al. 2005; Bartholomeusz and Locarnini 2006; Tersago et al. 2009, 2011; Muylaert et al. 2019)
<i>Viral meningitis</i>	Meningitis is considered a bacterial and viral disease. Viral meningitis can infect the human brain, and its infection can cause headaches, encephalitis, malaise, long-term sleep disorders, and depression. In most cases, there are no major side effects.	(Peate 1999; Schmidt et al. 2006; Tuppeny 2013; McGill et al. 2017)
<i>Mumps virus</i>	Mumps is a mildly infectious disease that affects children. However, in some situations, its systemic infection can have serious clinical effects, particularly when the sickness is postponed into adulthood. It infects pancreatic Beta cells, which can lead to fever, swelling of the parotid glands, fatigue, low-grade fever, attacks of tachycardia, and night sweating.	(Parkkonen et al. 1992; Gut et al. 1995; Nöjd et al. 2001; Mühlemann 2004)

170

171

172

Viruses such as the coronavirus (i.e., SARS-CoV-2) are intracellular parasites that are very small in the range of 20 to 400 nm (Yang Z., 2017). These viruses are resistant to

173 indoor air for a long time, especially in crowded or small environments. Inhaling these
174 viruses causes illness or worsens infections of the upper respiratory tract (Prussin et al.
175 2015). In the event of a viral pandemic, many countries implement containment measures
176 to prevent the spread of the virus. Still, this practice induces poor indoor air quality, which
177 will lead to other health problems. Indoor air in enclosed public spaces is at increased risk
178 of viruses because IAQ regulations are not adapted to current scientific research. So,
179 viruses can spread through the air and even easily enter indoor air (WHO, 2000). Once
180 locked in, even simple actions like talking, sneezing, and coughing that cause small virus-
181 filled droplets of saliva to spread through the air and onto surfaces, especially on the hands
182 (Alshraiedeh et al., 2013). Viral infection can spread from person to person, making
183 humans a significant source of indoor viral infection. Pets are also a foremost source of
184 indoor air contamination. Viruses can infect all living cell-based organisms, from bacteria
185 to humans, animals, and plants (Filipić et al., 2020).

186 Indoor air humidity is a major determinant of indoor air quality (Sakudo et al., 2014).
187 It plays an important role in viral infections by promoting the survival rate of viruses and an
188 increased risk of respiratory infections and allergic diseases (Aboubakr et al., 2018).
189 Several studies demonstrated that indoor temperature and humidity (generally between 19
190 and 26 ° C and 50 % - 60 % RH) positively affect the spread of infections and the
191 vulnerability of individuals to viruses. (Liang et al., 2020).

192 Pandemics such as COVID-19 require implementing new indoor air purification
193 techniques (Kim and Jang, 2018), replacing, combining, or improving existing conventional
194 viral inactivation methods. Conventional methods for viral inactivation, such as chlorine
195 disinfection, don't have an effective disinfection ability against viruses (Ashraf et al., 2013).
196 Unlike those methods, NTPs can deal with both transport and infectivity of airborne viruses
197 by filtration and interact with interactive plasma species (Xia et al., 2019).

198 **3. Parameters controlling viruses' inactivation performance**

199 Several parameters remain essential in the non-thermal discharge viruses'
200 inactivation, such as virus concentration, input power, air humidity, air composition, reactor
201 design, and type of discharge. It is also worth noting the oxygen concentration.

202 **3.1. Effect of the viruses' concentration**

203 The virus concentration effect in the nonthermal discharge viruses' inactivation was
204 reported in several studies (Chen et al., 2010). Indeed, Puligundla and Mok (Puligundla
205 and Mok, 2016) revealed a reduction of at least 4 to 6 log₁₀ of two bacteriophages at
206 concentrations of 10⁶ PFU/mL⁻¹ after 10 min of exposure to NTP.

207 After a couple of seconds of discharge treatment, the number of infectious phages
208 decreased quickly, and 6-orders of magnitude inactivation were achieved. On the other
209 hand, the virus concentrations used in NTPs ranked from 1x 10⁶ PFU mL⁻¹ to 300 PFU mL⁻¹
210 within several studies (Chen et al., 2010; Puligundla and Mok 2016; Jiang et al. 2017;
211 Filipić et al., 2020a). We noted 10⁵ TCID₅₀ /mL of bacteriophage MS2, matching 69 x 10⁶
212 PFU/mL according to Xia et al. (Xia et al., 2019) study, while that of Venezia et al.
213 (Alshraiedeh et al., 2013) denoted 10⁹ PFU/mL.

214 Thus, some works reported a proportional relationship between virus concentration
215 and the inactivation performance with nonthermal plasma: the inactivation yield of virus
216 decrease with very polluted indoor air. This finding might be due to the higher virus
217 concentration, and the step limitation is the reaction between reactive species and viruses
218 (Lee et al., 2021).

219 **3.2. Input power**

220 Input power is an important factor in influencing virus removal in NTP reactors. In fact, virus
221 elimination measurements are a function of input power (Mustafa et al., 2018). Several
222 previous studies demonstrated that the removal efficiencies of harmful airborne
223 components increase with the NTP input power (Shimizu and Oda 1999; An et al., 2011;
224 Pinard et al, 2019; Xia et al., 2019). It is well known that the power of the plasma is
225 proportional to the voltage applied in the discharge zone. Indeed, the increase in electric
226 power pleads in favor of having an increase in electric current in the vicinity of the virus.
227 Since the virus is in direct contact with the plasma, the exposure of the virus to species
228 produced by short-lived plasma, including electrons, ions, radicals, and UV rays, is
229 significantly higher compared to other treatment modalities. Likewise, the virus will also be
230 subjected to high electric fields at the plasma-surface interface. All of these active species
231 (OH°, O°, O₃, e⁻) ensure the inactivation of the virus (Mohamed et al., 2021).

232 Indeed, Xia et al. (Xia et al., 2019) examined viruses' inactivation through NTP coupled
233 with a packed-bed dielectric barrier discharge plasma. After NTP treatment, the results

234 revealed a higher inactivation of phage MS2 with a rise in the applied voltage up to 30 kV
235 (Alshraiedeh et al., 2013).

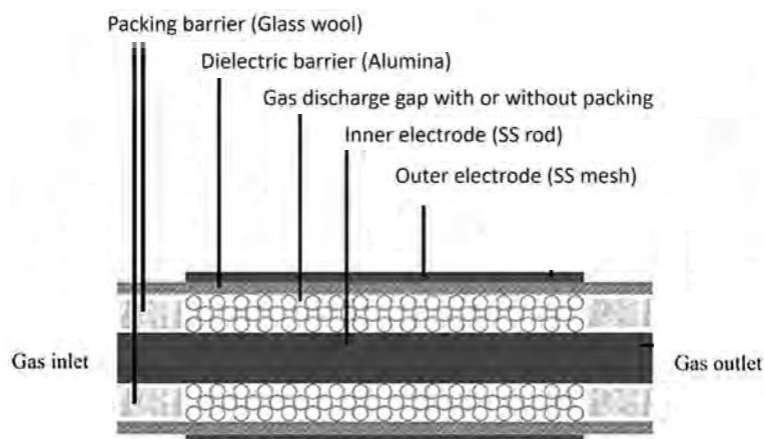
236 **3.3. Air humidity**

237 The nonthermal discharge viruses' inactivation can be affected by many
238 environmental factors such as air humidity versus the different reactors. For instance,
239 Cutler et al. (Cutler et al., 2012) demonstrated that for any temperature, the rate of the
240 inactivation of the airborne porcine reproductive and respiratory syndrome (PRRS) was
241 highest at a relative humidity between 25 % and 79 % and lowest at relative humidity > 80
242 %. Likewise, Xia et al. (Xia et al., 2020) found 98.6 % as the PRRSv inactivation efficiency
243 with 38 % of relative humidity under 20 kV voltage supplied to the packed bed NTP reactor.
244 However, some authors investigated the spread of relative humidity (RH) within the NTP
245 reactor, and they found that the RH evolved to reach 60 % at a steady state in 30 minutes
246 (Xia et al., 2019). This result obtained with the NTP reactor corroborates that of Cutler et
247 al. (Cutler et al., 2012) in the case of the PRRS virus, regarding the RH effect on the reactor
248 efficiency. On the other hand, other studies have reported that the relative humidity of the
249 PRRS virus depends on the viral envelopes (Elazhary and Derbyshire, 1979). Viruses with
250 lipoprotein envelopes tend to be more stable at lower relative humidity and non-enveloped
251 viruses more stable at higher relative humidity. Overall, relative humidity plays an important
252 role in the different reactors' yields. The divergence in the resulting level is due to the
253 difference in generating the plasma process, the residence time, reactor configuration, etc.

254 255 **3.4. Reactor for the inactivation of viral aerosols**

256 Over the past 20 years, several technologies have been devised for the inactivation
257 of non-thermal discharge viruses (Jacobs et al., 2010; Alshraiedeh et al., 2013; Sakudo et
258 al., 2014).

259 Recently, some studies examined microbial inactivation, sterilization, and disinfection
260 using APNTP within a field of biomedical application (Fridman et al., 2008). APNTP
261 applying possesses potential favor over standard chemical sterilants and disinfectants
262 (Figure 1). These include the design's simplicity and operation. It uses non-toxic gases with
263 the absence of toxic residues and the production of a large amount of various microbicidal
264 active species, including chemically reactive species (Yardimci and Setlow, 2010; Nayak
265 et al., 2018).



266
 267 Figure 1: Schematic of the DBD packed-bed reactor for the inactivation of viral aerosols
 268 (Michielsen et al., 2017; Uytendhouwen et al., 2018).

269
 270 Table 2: plasma reactor configuration for viruses and bacterial inactivation treatment for
 271 some types of the virus with reactive species

Virus	Reactive species responsible for the inactivation	NTP Reactor configuration	Mode of inactivation	References
SARS-CoV-2	RONS	Cylindrical Cold atmospheric plasma reactor (CCAPR)	DNA/RNA degradation	(Chen et al., 2020)
MS2 Bacteriophage	O	Tubular atmospheric-pressure cold plasma (TAPCP)	Both protein and DNA/RNA degradation	(Wu et al., 2015)
	O ₃		NA	(Xia et al., 2019)
Adenovirus	H ₂ O ₂	surface micro-discharge (SMD) plasma electrode using an insulator plate made of Al ₂ O ₃	DNA/RNA degradation	(Sakudo et al., 2016)
	O ₃		NA	(Zimmermann et al., 2011)

T4, Φ 174, and MS2	O ₂ , NO ₂ , and ONOO ⁻	Mesh electrode with a planar configuration	Both protein and DNA/RNA degradation	(Guo et al., 2018)
Feline CaliciVirus (FCV)	O and ONOOH	Tubular reactor plasma	Protein degradation	(Aboubakr et al., 2016)
	O ₂ and ONOO ⁻		DNA/RNA degradation	(Yamashiro et al., 2018)
	O ₂ and O ₃		Both protein and RNA degradation	(Aboubakr et al., 2018)
	NO _x and O ₃		NA	(Nayak et al., 2018)
Respiratory Syncytial Virus (RSV)	H ₂ O ₂	Tubular NTP reactor	DNA/RNA degradation	(Sakudo et al., 2017)
HIV-1	O ₂ ⁺ , O, NO and N ₂	Cylindrical Cold atmospheric plasma reactor (CCAPR)	DNA/RNA degradation	(Volotskova et al., 2016)
Newcastle Disease Virus (NDV)	H ₂ O ₂ , OH [*] and NO _x	Magnetically Rotated Gliding Arc & DBD plasma planar reactor	Both protein and DNA/RNA degradation	(Su et al., 2018)
	RONS		RNA degradation	(Schiappacasse et al., 2020)
Influenza viruses	H ₂ O ₂		Both protein and RNA degradation	(Sakudo et al., 2014)
Influenza A	OH and O ₃		NA	(Gallagher et al., 2004)

272

273 4. Combined system for virus disinfection

274 Several works have been reported on conventional techniques for viruses and
275 bacterial inactivation treatment, whether physical or chemical (table 3). Some technologies
276 have been combined with different forms of electric discharge plasma or other
277 decontamination processes to deal with various pathogenic microorganisms' high levels of

278 contamination to improve inactivation efficiency (Bourke et al., 2018; Kordová et al., 2018;
 279 Rtimi and Kiwi, 2021).

280 **Table 3:** Summary of studies on plasma combination technologies for virus inactivation.

Combination systems	Virus types	Experimental conditions	Results	Ref.
Non-thermal plasma-activated by NaCl or H ₂ O ₂ solutions	Newcastle Disease Virus (NDV)	10 mL sterile distilled water, 0.9 % NaCl, and 0.3 % H ₂ O ₂ solutions	Complete inactivation of NDV after only 30 min of treatment	(Su et al., 2018)
Atmospheric pressure non-thermal plasma (NTP) jet with Ar/O ₂ /N ₂	Newcastle Disease virus (NDV) strain and H ₉ N ₂ avian influenza virus (AIV)	A mixture containing 88 % Ar, 2 % O ₂ and 10 % N ₂ at a flow rate of 5 L/min was used as working gas	Complete inactivation in 2 min of treatment for both NDV and AIV	(Wang et al., 2016)
Cold oxygen plasma + an internal classic UV-C lamp	Respiratory syncytial virus (RSV) and human parainfluenza virus type 3 (hPIV-3), and influenza virus A (H ₅ N ₂)	50 mL of Viral pellets were suspended in PBS	More than 99.98 of reduction regardless of the virus type	(Assadi et al., 2015)
Non-thermal plasma coupled to a packed-bed dielectric barrier discharge reactor	Aerosols of phage MS2	At 30 kV and an airflow rate of 170 standard liters per minute	A reduction of ~2 log of the MS2 inactivated and ~0.35 log physically removed in the packed bed was observed	(Xia et al., 2020)
packed-bed dielectric barrier discharge non-thermal plasma combined with Filtration	porcine reproductive and respiratory syndrome virus (PRRSv)	Form 12 to 30 kV With RH = 38 -49 % at 5 -12 cfm	More than 40% at 12 cfm and more than 80% at 5 cfm of reduction regardless of the virus SARS (MS2)	(Xia et al., 2019)

281 In recent years, current trends have been moving towards the coupling of plasma and
 282 nanomaterials with dimensions in the order of a nanometer (i.e., 1 to 100 nm). This process

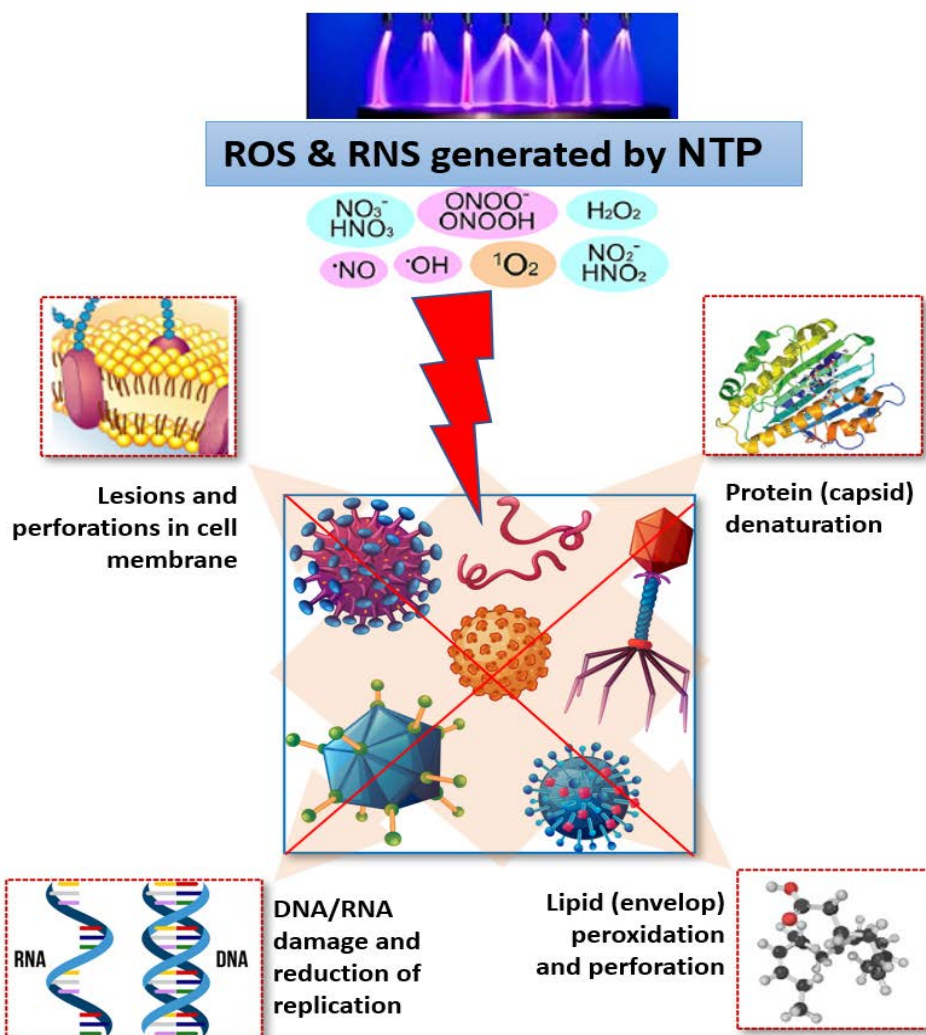
283 has been well confirmed with the inactivation effects of nanomaterials on various viruses
284 due to a large specific surface area, including the generation of ROS (Abidi et al., 2019,
285 2020). The effects resulting from the combination between the plasma in the catalytic
286 oxidation, the photothermal influences, and the release of metal ions allows having a
287 synergistic impact on the antiviral performances (Li et al., 2021). Therefore, there is a
288 comprehensive promise in developing antiviral nanomaterials and using them to air purifier
289 filters, building ventilation systems, antiviral fabrics, and antiviral spray agents, which
290 assists in controlling the spread of viruses through aerosols.

291 **5. Mechanism of viral inactivation by the non-thermal discharge**

292 Nonthermal plasma constitutes one of the recent processes of Advanced Oxidation
293 Processes (AOPs). This technology allows the formation of heavy mixtures and lighting
294 from the excitation of gases by electric discharge. Probably, the effectiveness of NTPs is
295 because they combine both physical and chemical mechanisms (Ateia et al., 2020).
296 However, these coupled techniques generate highly reactive oxidizing components, free
297 radicals, and ions (e.g., ozone, hydrogen peroxide, hydroxyl, and superoxide radicals) in
298 aqueous solutions under UV irradiation (Wang et al., 2018). Figure 1 shows a schematic
299 of the action of NTP of viral inactivation by the non-thermal discharge. The generation of
300 reactive oxygen species (ROS) and/or reactive nitrogen species (RNS) is the main feature
301 of NTP that contributes to virus inactivation. The main target of ROS/RNS is virus capsid,
302 genome, protein, and glycoprotein. The short lifespan of ROS or RNS makes the
303 identification and assessment of ROS/RNS a challenging task. Furthermore, the
304 experimental conditions (the gas used for the NTP generation, the matrix, the virus
305 treated...etc.) play a crucial role in the generation of particular ROS/RNS.

306 The most crucial component for evaluating virus inactivation is the degradation of the
307 capsid. The viral envelope or capsid is the first contact point with the host cell. For an
308 efficient recognition of a virus by the cell receptors, their outer structure must be intact.
309 The increased oxidative force of NTP can denature the virus and genome structure by
310 affecting both proteins and nucleic acids. Conformational disruption of the capsid proteins
311 caused by ROS/RNS can result in loss of viral infectivity due to the virus's disruption
312 binding to receptors on the host cell membrane (Filipić et al., 2020b).
313 In addition to capsid and nucleic acid, further damage could be seen in lipid components
314 from the envelope the virus influenza A (Sakudo et al., 2014). Particularly in the case of
315 bacteriophage λ (Yasuda et al., 2010) and FCV (Aboubakr et al., 2018), it has been proved

316 that the main mode of NCP inactivation is the degradation of the capsid proteins
 317 proceeding with the degradation of nucleic acids (figure 2). However, in several studies, it
 318 was impossible to determine which degradation path contributed more to the decay in viral
 319 infectivity (Filipić et al., 2020b).
 320

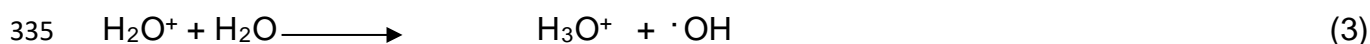
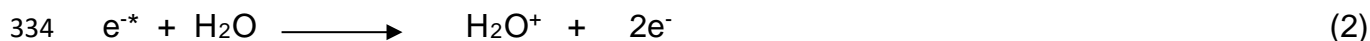
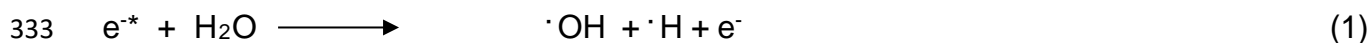


321
 322 **Figure 2:** A schematic of the action of NTP of viral inactivation by the non-thermal
 323 discharge.

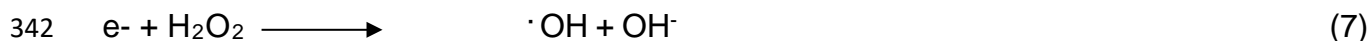
324 **5.1. Chemical processes' reactions**

325 It is worthy to noted that in humid air, electrons with very high kinetic energy and could
 326 react with water vapor to occur reactive species. Then, some reactive chemical species
 327 (*i.e.*, H₂O₂, O₃, O₂⁻, ·OH) also react by attacking the high-energy electron (Wang et al.,
 328 2018). In fact, electrons energy generated through the electrical discharge plasma process
 329 can outdo the decay or ionization energy of water molecules (Joshi et al., 1995; Wang et

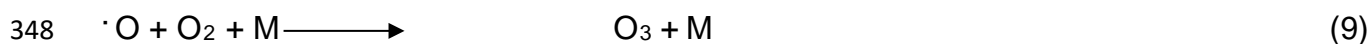
330 al., 2008). Through electron collisions, the dissociation of water molecules in the air could
331 form $\cdot\text{H}$, $\cdot\text{OH}$, and other hydrated cations as follows (Hong and Zeng, 2002; Wang et al.,
332 2008):



336 Where * denoted high-energy electron state. These formed radicals could react together
337 or recombine with other reactive species, as presented in the following reactions (Zhou et
338 al. 2016):



344 Despite the free radicals' generation for virus degradation in NTPs, ozone (O_3) reactions
345 also occur. Once $\cdot\text{O}$ is formed (*i.e.*, equation 8) *via* high-energy electronic attack, it could
346 react with O_2 and organic matter (M) to generate O_3 , as shown in the reaction (9) (Wang
347 et al., 2018).



349 Where M is organic matter, O_3 could react with various compounds through direct O_3 and
350 indirect O_3 oxidation reactions. In previous studies, also it was found that the ozone
351 produced by plasma was not the main contributor to the inactivation effect; nevertheless,
352 it is involved in the inactivation (Morrison et al., 2021). In fact, Morisson and her co-workers
353 (Morisson et al., 2021) showed that ozone disinfection had demonstrated high efficacy
354 against enveloped and non-enveloped viruses, including viruses similar in morphology to
355 SARS-CoV-2.

356 **5.2. Physical processes' reactions**

357 The physical processes of the inactivation effect in non-thermal plasma can occur *via* UV
358 irradiation (Sun et al., 2005; Wang et al., 2006; Tang et al., 2018), shock waves (Šunka
359 2001; Ghernaout and Elboughdiri, 2020), and electric field effects (Schoenbach et al.,
360 1997, 2000). Indeed, the UV irradiation from gas-phase discharges is weaker than that
361 from the liquid phase. The types of UV irradiation from gas-phase discharges at
362 atmospheric pressure are generally within the UVA and UVB regions, with wavelengths
363 ranging of 320–400 nm and 280–320 nm, respectively. Matsumoto et al. (Matsumoto et al.
364 1992) reported that UV irradiation performed an essential part in bacteria inactivation.
365 Moreover, UV irradiation has also been used in the gas phase discharge plasma process
366 (Xue et al., 2008).

367 In the absence of electric discharge, the electric field is an alternative method for
368 inactivating cells and processing food products. Previous research reported that a potent
369 electric field could inactivate the microorganisms and biofilm bacteria on the walls of
370 cooling and drinking water pipes (Schoenbach et al., 2000).

371

372 **6. Conclusions and Outlook**

373 Over the past decades, interest in applying nonthermal plasma technology for virus
374 inactivation within indoor air has considerably increased. In this review paper, commonly
375 used nonthermal plasma reactors and their effectiveness in virus inactivation have been
376 extensively exhibited. Moreover, the inactivation mechanisms and the factors controlling
377 these reactors, such as virus concentration, input power, air humidity, air composition, and
378 reactor design, have also earned attention.

379 Notwithstanding the determining factors in nonthermal plasma processes, the
380 production and evolution of active species and the physical effects of viral inactivation must
381 be further investigated to explain the mechanisms of plasma oxidation and optimize the
382 oxidation nonthermal plasmas process. Furthermore, investigations should be performed
383 on the impact of target properties, including volatility, hydrophobic/hydrophilic nature,
384 stability, and diffusivity. However, nonthermal plasma can be combined with other
385 technologies involving catalysts like carbonaceous materials, metal oxides, metal ions, etc.
386 The mechanism of interaction between viral inactivation and catalysts needs to be well

387 studied. Well-mastered nonthermal plasma could be a promising alternative to face these
388 waves of COVID-19 infections.

389

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