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Airborne contamination in the food industry: An update on monitoring and disinfection techniques of air

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## ABSTRACT

*Background:* Hygienic and safe production is a high priority in the food industry. During processing, food may be subjected to bio-contamination. Accordingly, preservation of overall quality by keeping a clean environment is a goal to pursue. Among microbial vectors, air is considered a contributing factor to cross-contamination.

*Scope and approach:* Nowadays, in food plants emphasis is paid to the assessment of air bioload in view of prevention of recontamination. Normally, air entering a processing plant is chilled and filtered to remove undesired microorganisms from outside. Nevertheless, apart from clean-room environments, uncontrolled factors (processes, personnel, structures, etc.) contribute to the release of microorganisms in indoor environments, resulting in generation of bioaerosols highly variable within and among plants, and on a daily basis within the same plant.

*Key findings and conclusions:* This review focuses on the relevance of bioaerosol monitoring in the food industry, providing an update of air sampling techniques and methods of analysis in view to strengthen preventive hygienic actions. Disinfection procedures to minimize microbial counts in the air as additional safeguard to the standard chemical sanitation protocols are reviewed. Benefits and limitations of air treatment by chemical fogging, ozonation, UV irradiation or cold plasma are outlined. Air bioload monitoring and the implementation of subsequent air disinfection procedures are a feasible and a routinely exploitable strategy to satisfy hygienic requirements in food plants. Further research is required to face technical challenges and optimize the feasibility of some disinfection technologies for the real-world of food environments.

1 **AIRBORNE CONTAMINATION IN THE FOOD INDUSTRY: AN UPDATE ON MONITORING**  
2 **AND DISINFECTION TECHNIQUES OF AIR**

3

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5

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30 world of food environments.

31

32 **Keywords:** Bioaerosol; Air monitoring; Chemical fogging; Gaseous ozonation; UV irradiation.

33

### 34 **Microbial contamination of food: routes, vectors and factors limiting spreading**

35 Food contaminants are classified as extraneous substances of either physical, chemical or biological  
36 origin. Microorganisms may be responsible for outbreaks of food-related illnesses or food spoilage. In a  
37 generic food facility, major routes of food recontamination by microorganisms are via surface contact, via  
38 personnel or via the air (Figure 1) (den Aantrekker, Boom, Zwietering, & van Schothorst, 2003).  
39 Generally, the contribution of the first two routes is prevailing, but the importance of each means of  
40 contamination is also a function of the type of product or process. This review deals with items related to  
41 food contamination by air route. Employees can transfer microorganisms both directly (from their body to  
42 the food product) and indirectly (transferring contamination from one area/surface to another) (Aarnisalo,  
43 2007). In this context, the Annex II of the European Regulation No 852/2004 on food hygiene (EC, 2004)  
44 takes into consideration the relevant role of employees, establishing their supervision and instruction in  
45 food hygiene matters in relation to the work activity. Also exposure to contaminated surfaces has been  
46 identified as a major source of food contamination (Otto et al., 2011). Both food-contact (e.g., equipment,  
47 utensils, workbenches, conveyor belts) and no food-contact surfaces (e.g., drains, utility pipes,  
48 maintenance equipment, structures, and areas away from production such as hallways, entrances and  
49 welfare facilities) can collect microorganisms and other debris from employees, as well as from the air  
50 and other materials. These mutual interactions among above cited vectors can boost the microbial spread  
51 in a food facility (Figure 1). In general, the low incidence and/or viability of pathogens in suspension in  
52 the air makes the route of air-to-food of low impact on foodborne diseases (Pérez-Rodríguez, Valero,

53 Carrasco, García, & Zurera, 2008). Nonetheless, the recontamination by air is noteworthy for products  
54 such as beverages, refrigerated dairy and culinary products and products with very low viable counts,  
55 such as dried infant formulae (Reij, den Aantrekker, & ILSI Europe Risk Analysis in Microbiology,  
56 2004). In high-risk areas, for instance after the last heat treatment before filling and packaging, the food  
57 product (e.g., beverages) is susceptible to recontamination. In dairy production facilities, spray drying and  
58 milling operations have been reported as a possible means of microbial transfer, making dissemination of  
59 pathogens through ventilation a probable event (Mullane, Whyte, Wall, Quinn, & Fanning, 2008). To  
60 counteract the risk of airborne biocontamination in the filling room, air filters should be changed on a  
61 regular basis, and a positive air pressure should be adopted (Lawlor, Schuman, Simpson, & Taormina,  
62 2009). By modelling studies, den Aantrekker et al. (2003) carried out a quantitative estimation of the  
63 probability of product contamination via the air. Assuming settling velocities of microorganisms under  
64 the influence of gravity only, the authors took into consideration what-if scenarios to exemplify the  
65 determination of design criteria to control a specified contamination level. As a conclusion, both the type  
66 of product and processing conditions strongly influence the contamination level. Comprehensive  
67 approaches to model factory air movements have been described in literature and represent a contribution  
68 of research to improve the understanding and tackling of microbiological risks (Pérez-Rodríguez et al.,  
69 2008; Possas, Carrasco, García-Gimeno & Valero, 2017).

70 Other factors can contribute to microbial transfer to food, namely, raw materials, ingredients, pests,  
71 water, processing conditions, packaging material, transport vehicles, plant design, poor zoning, open  
72 drains, as well as wet and dry cleaning operations by brushing, which often result in the generation of  
73 bioaerosols in the form of water droplets or dry dust (Ehaval, 2007; Marriott & Gravani, 2006). If  
74 cleaning and disinfection procedures are not performed in the correct manner, residues of organic and  
75 inorganic soils could remain, and subsequently food spoilage and pathogenic bacteria could create a  
76 suitable environment for biofilm development. In a wide range of food industries, biofilms have become  
77 challenging (Marino, Maifreni, Baggio, & Innocente, 2018). In the topmost layers of the biofilm, chunks  
78 of the extracellular polymeric substances, with the accompanying microbial population, can cross-

79 contaminate other products, by the action of food or liquid passing over the surface (Marriott & Gravani,  
80 2006). To the best of our knowledge, to date, detaching and air diffusion of above-mentioned substances  
81 have not been reported.

82 Generally, epidemiological data on common contamination routes and sources are scarcely described  
83 in the literature (Reij et al., 2004). Recent research in this area is focused to achieve greater insight into  
84 the mechanisms of microbial transfer and cross-contamination dynamics during food processing (Possas  
85 et al., 2017). Considering the complexity of parameters involved in microbial transfer, it is apparent that  
86 only an integrated approach may be effective to prevent or minimize food contamination. Hygienic design  
87 of equipment/structures and proper sanitation are factors limiting the microbial contamination in full  
88 compliance with legislation (EC, 2004; EN 1672-2, 1997). Good hygiene practices include also personal  
89 hygiene, zone separation, prevention of cross-contamination, use of purified water (Gurnari, 2015).  
90 Additional actions in the management of food processing such as proper selection of ingredients, food  
91 storage conditions, plant maintenance and air filtration are efficient tools in view of keeping or improving  
92 food safety. The relative contribution of these factors is variable as a function of the food sector.

93 Food hygiene is currently defined as measures and conditions necessary to control hazards and ensure  
94 the safety of food at all stages of the chain (Codex Alimentarius, 2003; EC, 2004). It is realized through  
95 established prerequisite programs, including good manufacturing practices (GMP), good hygiene  
96 practices (GHP) and standard operating procedures (SOP), which contribute to make hazard analysis  
97 critical control point (HACCP) an effective system to control food safety (Byrne, Lyng, Dunne, & Bolton,  
98 2008; Varzakas, 2016). Even with the best control measures in place, a food product may still pose a risk  
99 to the consumer (den Aantrekker et al., 2003). Thus, all means to reduce or prevent contamination and to  
100 improve the suitability for consumption are considered part of the hygiene concept. A proper management  
101 of air quality can mitigate the introduction of microorganisms throughout the production stream of a food  
102 product. Each food production facility should evaluate the presence of microorganisms in the site,  
103 sampling both surfaces and the air, through the implementation of an environmental monitoring program  
104 (EMP) necessary for the subsequent development of a food safety plan (FPS) (Pleitner, 2018). The

105 developed EMP allows to evaluate the effectiveness of the microbial controls in place. Such activity is  
106 pivotal in a well-run company.

107 This review aims at highlighting the role of the airborne route in the microbial spreading in the food  
108 industry. The scope is to provide an overview on both bioaerosol monitoring, including air sampling  
109 techniques and methods of analysis, and on subsequent air disinfection procedures as a proactive strategy  
110 in addition to routine sanitation practices. The items covered in this review are addressed to food safety  
111 aspects. Studies related to the field of occupational health are outside of the scope. The major target  
112 readers are food business operators who can perceive the potential advantages in terms of food safety  
113 arising from the implementation of environmental control protocols.

114

#### 115 **What is a bioaerosol and why air monitoring is important?**

116 The suspensions of microscopic solid or liquid particles in the air are defined as aerosols (Ferguson,  
117 Cumbrell, & Whitby, 2019). Those of major impact in the food sector are known as bioaerosols and  
118 consist of living substances with diameters up to 50  $\mu\text{m}$  (Burfoot, 2016). These may include bacteria,  
119 mold spores and yeasts (Lee, 2011). Indeed, although rarely documented, phage contamination can also  
120 occur through aerosolization (Verreault et al., 2011). Viruses can be found on aerosol particles of various  
121 sizes, from the submicrometer range to tens of micrometers in aerodynamic diameter. Virtually all  
122 microorganisms present in bioaerosols are easily translocated by air currents, but their reproduction is  
123 uncommon in the air due to the lack of moisture and nutrients. Despite the sensitiveness to environmental  
124 conditions, also food pathogens can survive in the air, for instance in association with dust particles  
125 (Mullane et al., 2007). Additionally, contamination from airborne yeasts and molds can affect the quality  
126 and shelf life of a food product (Ehaval, 2007). The bioaerosol of the food industry is a mixture of many  
127 species of microorganisms including bacteria endospores and exospores (e.g., *Bacillus*, *Clostridium*),  
128 vegetative cells mainly of Gram positive bacteria (e.g., *Micrococcus*, *Staphylococcus*), molds (e.g.,  
129 *Penicillium*, *Cladosporium*, *Alternaria*, *Fusarium*) as well as yeasts (e.g., *Saccharomyces*, *Torulasporea*,  
130 *Hanseniaspora*, *Pichia*) (Pérez-Martín, Seseña, Fernández-González, Arévalo, & Llanos Palop, 2014).

131 Aerosolized microorganisms may persist within droplets derived from the aerosolization of water  
132 spraying/splashing during food processing or the sanitation process. In these cases, microorganisms grow  
133 in a liquid medium, such as spilled product, rinse water or wastewater, which subsequently becomes  
134 aerosolized. Microorganisms may also be suspended as such in the air after dissipation or evaporation or  
135 as “passengers” on solid dust particles (e.g., hair, clothing fiber, skin), which are dispersed in a food  
136 processing unit (Chang, Ting, & Horng, 2019; Heo, Lim, Kee, & Lee, 2017). Microorganisms in the air  
137 may settle on food products, equipment, containers and other food contact surfaces during handling  
138 (Brandl et al., 2014). Any point at which the food product is exposed to air is a possible route for airborne  
139 contamination. Combining the knowledge acquired under real situations in food factories and the use of  
140 computer models, Burfoot (2016) reported that the smaller the particle suspended in the air the greater the  
141 flight time and the distance it may travel. Indeed, the fate of airborne particles is quite complex and ruled  
142 by several mechanisms including: gravitational settling, Brownian diffusion, inertial impaction, direct  
143 interception (by, for example, van der Waal’s forces) and electrostatic attraction (Da, Géhin, Havet,  
144 Othmane, & Sollicec, 2015). The combination of above-mentioned parameters influences the aerodynamic  
145 behavior of particles affecting the success of the air sampling. Generally, the airborne particles most of  
146 interest in food environments are those containing bacteria with low-medium size (above 1  $\mu\text{m}$  and below  
147 20  $\mu\text{m}$ ) which can disperse easily around the generation area. By the way, aerosols in food plants have not  
148 been studied sufficiently to accurately generalize particle-size distribution. Generally, in high-care areas  
149 less than 1 % of particles in the air will settle, and most of them will be removed by the filtration system.  
150 The contribution of airborne microorganisms to food contamination has been addressed (Chang, et al.,  
151 2019; Chen et al., 2019; Shale & Lues, 2007). Burfoot and Brown (2004) reported that the ratio of  
152 microorganisms to total particles may range up to more than two orders of magnitude. For instance, these  
153 authors observed in different food factory environments that above-mentioned ratio was low (1 to 30,000)  
154 in periods of inactivity in a well-designed production area, whereas it reached high levels (about 1 to 200)  
155 near to employees during hand-washing and next to cleaning operations. To date, the awareness of the  
156 industry about the importance of the hygienic design, remarkably for the air handling system, is still low



157 (Da et al., 2015). Nonetheless, overemphasis on the role of air as a source of food contamination should  
158 be avoided. Burfoot, Whyte, Tinker, Hall and Allen (2007) quantified the contribution of airborne  
159 microorganisms to contamination of poultry carcasses undergoing processing in an evisceration room.  
160 The use of ultra-clean air provided by a high-efficiency particulate air (HEPA) unit reduced total aerobic  
161 counts on horizontal settle plates by 68-fold. Differently, after measurement by sponging, the use of ultra-  
162 clean air had no effect on the counts on carcasses. The latter resulted so heavily contaminated that the  
163 airborne bacteria in the evisceration room represented less than 1 % of total number of bacteria on  
164 carcasses.

165 The food industry is aware that monitoring aerosols is becoming a must in standard quality-control  
166 practices. Generally, the primary focus is addressed to total viable microorganisms rather than total  
167 particle counts. Air monitoring can be included as a part of an HACCP system in the food industry  
168 (Beletsiotis, Ghikas, & Kalantzi, 2011). The role of bioaerosol monitoring consists in:

- 169 - being the basic step for prevention;
- 170 - implementing a pro-active action to minimize cross-contamination phenomena, which are major  
171 contributors in food-borne outbreaks;
- 172 - complying with legal requirements or guidelines stating that the air in food sector has to be controlled  
173 without specifying the methodology or minimum acceptable standards (Wray, 2011);
- 174 - finding the potential source of new contamination whenever any structural implementation has been  
175 introduced, and subsequently undertaking appropriate corrective measures;
- 176 - collecting epidemiological data, possibly with a view to set occupational exposure limits (Wirtanen,  
177 Miettinen, Pahkala, Enbom, & Vanne, 2002).

178 Information sources provided by the food legislator are quite generic. The European regulation states  
179 the need to minimize airborne contamination and to avoid mechanical airflow from a contaminated area  
180 to a clean area (EC, 2004). Guidances, intended to assist food producers to meet the air quality and  
181 hygienic requirements of the food manufacturing process, are available. The European Hygienic

182 Engineering and Design Group (EHEDG) supported the European legislation producing a guideline  
183 focusing on air handling systems installed in the food industry for air quality control (EHEDG, 2016).

184

#### 185 **Bioaerosol monitoring: air sampling techniques and methods of analysis**

186 The assessment of air microbial load in the food industry is performed through the sampling of a  
187 representative amount of air and its subsequent analysis. Quantification and identification of bioaerosols  
188 is affected by several factors, such as the rate with which the result is required, the efficiency of sampling  
189 equipment, the ratio of total cell counts versus viability of cells in the sample, the particle size range  
190 selected as well as the analysis methods (Dybwad, Skogan, & Blatny, 2014). Once the reasons for  
191 carrying out sampling have been defined, the rate of relevance of above-mentioned parameters can be  
192 established. The samplers should apply minimum stress during air collection to reduce the impairment of  
193 the biological activity of the aerosol. In addition, during air sampling, different environmental parameters  
194 can cumulatively stress microorganisms affecting (through desiccation) their viability. In long-term (> 30  
195 min) sampling of bioaerosols, especially for vegetative bacteria, the combination of controlled humidity  
196 and refrigerated temperature of air sampler should provide viability maintenance (Walls et al., 2017). The  
197 literature provides little information on the causative variables that lead to differing colony recoveries  
198 (Wirtanen et al., 2002). Through years, to monitor air in a consistent way, performance measurements for  
199 air samplers have been reported using several efficiency terms, including aspiration-, sampling-,  
200 recovery- and overall-efficiency (Dybwad et al., 2014). The sampler efficiency is described also by  
201 factors such as the design of the inlet, collection stage and choice of collection medium, which affect the  
202 viability of microorganisms. Generally, the collection efficiency is expressed as the 50% aerodynamic  
203 cut-off diameter,  $D_{ae50}$  ( $\mu\text{m}$ ), i.e. the particle size collected to 50% diameter. The proper choice of a  
204 sampler with a  $D_{ae50}$  below the mean size of the particles being sampled is crucial for efficient collection.  
205 The performance information supplied with commercially available samplers is often limited to collection  
206 efficiencies, but data on sampling stress are not always provided. Summing up, the evaluation of air  
207 microbial load is not a trivial task. It can be performed through several sampling methods each with pros

208 and cons (Table 1). Recently, Reponen (2017) reviewed the techniques of air sampling of microorganisms  
209 in generic environments providing a list of commercially available bioaerosol samplers. Both passive  
210 (settle plates) and active (using a sampling device) air sampling techniques can be adopted (Haig,  
211 Mackay, Walker, & Williams, 2016; Reponen, 2017). The former approach consisting in the exposure of  
212 agar plates to air for a certain period of time has been traditionally used. In this case, the collection is  
213 governed by gravitational force, which is related to the particle mass. Settle plates technique is not  
214 quantitative, and in high aerosol concentrations the uncountable numbers of colonies may represent a  
215 problem. Active bioaerosol sampling exploits different collection principles, such as impaction,  
216 impingement, cyclonic separation, filtration, thermal or electrostatic precipitation. A large number of  
217 commercial samplers is available on the market. Nevertheless, different results are obtained from  
218 different equipment in the same place, at the same time (Verreault, Moineau, & Duchaine, 2011).  
219 Properties and critical factors affecting the use of air samplers have been recently reviewed by Brown and  
220 Wray (2014). Data comparison is difficult because the type of the device is reflected in the biodiversity of  
221 the bioaerosol (Mbareche, Veillette, Bilodeau and Duchaine, 2018). Dybwad et al. (2014) through a  
222 comparative evaluation of 9 different samplers (impactors, impingers, cyclones, electrostatic precipitators  
223 and filtration samplers) revealed significant differences in terms of cultivation-based biological sampling  
224 efficiencies and PCR-/microscopy-based physical sampling efficiencies as a function of the bioaerosol's  
225 stress-sensitivity and particle size. Typically, impaction is a common technique for the collection of  
226 airborne viable particles (Miettinen, 2016). In particular, there are two types of solid-surface impactors:  
227 slit samplers and sieve samplers, the latter being preferred. In a sieve sampler the air is drawn through a  
228 large number of small, evenly spaced holes drilled in a metal plate. Air particles impact on an agar surface  
229 located below the perforated plate. The Andersen sampler, a cascade-sieve impactor is likely the most-  
230 known device giving information on the size distribution of the microbiological aerosol. Liquid-using  
231 impactors, called impingers, are useful for sampling heavily contaminated air thanks to the dilution of the  
232 liquid sample for the subsequent culture growth analysis. Other instruments adopted in the food industry  
233 include centrifugal samplers based on cyclonic separation. In this case, air is pulled into the sampling unit

234 and pushed outside thus impacting on a strip of nutrient agar. Such device is characterized by selectivity  
235 for large particles, which are likely to include viable particles. For this reasons the tendency is to exhibit  
236 higher counts than with other devices. A further type of active sampler, relying on filtration as a  
237 collecting mechanism, is the filter system, which is recognized to be suitable for the subsequent  
238 enumeration of mold or bacterial spores. Airborne microorganisms can be collected also through  
239 electrostatic precipitators following ionization and subsequent deposition in an electric field on a growth  
240 medium. The adoption of this technique resulted more efficient than other methods (such as impingers)  
241 for sensitive microbial strains e.g., *Pseudomonas fluorescens* (Miettinen, 2016). Each of the above-  
242 mentioned devices has limitations that the user should be aware of. To date, in the food industry settle  
243 plates and impactors, being simple and practical, are the most used devices for routine microbial air  
244 monitoring.

245 After collection, the air sample is analyzed through culture, microscopic, biochemical, immunological or  
246 molecular assays (Mbareche, Brisebois, Veillette, & Duchaine, 2017; Reponen, Willeke, Grinshpun, &  
247 Nevalainen, 2011). The choice of the analytical method relies on factors including cost, time required,  
248 sensitivity, specificity and the sampling method used. The selection is defined before air sampling is  
249 carried out. Traditionally, in the food industry culture-based methods prevail for enumerating the airborne  
250 microbial counts (Oppliger, 2014). Microorganisms collected by impaction are cultured directly, whereas  
251 following the use of filter systems the transfer to a culture medium is required. Usually, for surveys on the  
252 characterization of the airborne microbiota the selection of general media is preferred, because it favors  
253 the growth of a large diversity of species. The simultaneous isolation of both bacteria and fungi is not  
254 satisfactory using only one culture medium. In case of volumetric samplings, the concentration of  
255 cultivable airborne microorganisms is obtained by referring the colony forming units (CFU) to the  
256 volume of air sampled. The limitation of plate count method is that it reveals only a part of the microbial  
257 population. Some bacteria may be in an eclipsed state defined as viable but not cultivable (VBNC) as a  
258 response to stress conditions (Maukonen, 2007). Despite this disadvantage, plate count method is by far  
259 the gold standard in food microbiology. In addition to culture technique, also microscopic analysis is used

260 to estimate the total number of microorganisms in an air sample, allowing enumeration of both cultivable  
261 and non-cultivable microorganisms. Direct microscopy is generally employed to identify fungi, exploiting  
262 the morphological characteristics of spores. Phase-contrast microscopy allows to count bacterial  
263 endospores due to their phase-bright appearance in contrast to darker vegetative cells. Recently,  
264 investigations focused on health effects following exposure to harmful bioaerosols, led to a demand for  
265 accurate and reliable monitoring systems (Choi, Kang & Jung, 2015). Molecular techniques such as  
266 polymerase chain reaction (PCR) amplification of 16 S rDNA, followed by its sequencing and DNA-  
267 DNA hybridization allow to increase sensitivity and specificity, while decreasing the time required for  
268 analysis (Stetzenbac, Buttner, & Cruz, 2004). Indeed, in the food industry, the development of real-time  
269 continuous monitoring of microorganisms in the air would be important to verify the occurrence of  
270 undesired trends that are not always revealed with periodic samplings. Through years, the quantitative  
271 PCR ((q)PCR) developed in the medical research area for assessing total or species-specific airborne  
272 bacterial load. Besides, the use of (q)PCR is more suitable than other techniques for the analysis of air  
273 samples in the detection of phage genome (Verreault et al., 2011). In this case, various sampling devices  
274 can be used to recover airborne viruses. Nevertheless, it is still challenging to study viral aerosols using  
275 metagenomics mainly due to limited quantity of viruses in the air samples and due to the limited viral  
276 databases for viral metagenome library analysis (Behzad, Gojobori and Mineta, 2015; Prussin, Marr and  
277 Bibby, 2014). To date, the most common techniques to recover viruses are liquid and solid impactors as  
278 well as filters. An extensive compilation of studies (mostly experimental in controlled chambers) on the  
279 recovery of viral particles was carried out by Verreault et al. (2011). The (q)PCR technique is advantaged  
280 by the coupling to other molecular methods (like sequencing and DNA-DNA hybridization) to obtain  
281 information about the species diversity (Oppliger, Charrière, Droz, & Rinsoz, 2008). The sensitivity of  
282 (q)PCR is of different orders of magnitude higher than that of culture techniques. Moreover, it is able to  
283 amplify the DNA of VBNC cells. Nonetheless, given current available technologies, it is impossible to  
284 real-time monitor all the airborne biological agents and classify them to the species level (Yao, 2018). To

285 date, in the food industry, despite the above discussed advantages, biochemical and molecular methods  
286 are not applied as routine techniques to monitor indoor microbial air quality.

287

### 288 **Levels of air contamination in commercial food processing plants**

289 The presence of microorganisms in the air of food facilities is predominantly accidental and is highly  
290 variable or transient, generally ranging from 10 to 10,000 CFU/m<sup>3</sup> (Ehaval, 2007). Based on the  
291 assumption that it is impossible to keep microbial counts at zero level, information on bioaerosol is  
292 important to evaluate the risk on both product quality and/or shelf life and public health.

293 In processing plants producing pork, poultry, beef and dairy products, air has been recognized as a  
294 contributor to food contamination. In particular, environments such as slaughterhouses are potentially  
295 critical, because animals are a microbial source of contamination. Prendergast, Daly, Sheridan, McDowell  
296 and Blair (2004) investigated the aerobiology of slaughter operations in two commercial beef abattoirs.  
297 Although quantitatively different, both of them showed a similar trend in counts within intraday  
298 processing, with lower levels before slaughtering (about 1 log<sub>10</sub> CFU/m<sup>3</sup> of air). The authors observed  
299 differences in the aerial contamination among different sites in one abattoir. In this case, total viable  
300 counts differed significantly ( $P < 0.001$ ) ranging from 1.79 up to 3.47 log<sub>10</sub> CFU/m<sup>3</sup> of air in the zone  
301 collecting washed carcass (“clean area”) and in the exsanguination site (“dirty area”), respectively. This  
302 pattern was not observed in the other abattoir due to the different building design, which allowed to  
303 effectively reducing the penetration of airborne contamination from “dirty” to “clean” areas. In addition  
304 to what has been already mentioned, Pearce, Sheridan and Bolton (2006) in a pork slaughtering plant  
305 enumerated about 1 log<sub>10</sub> cycle decrease of aerobic mesophilic bacteria from the “wet” room (bleeding  
306 site) to the “clean” room (chilling site). The authors pointed out the role of animals as a source of air  
307 contamination.

308 In a dairy plant, Beletsiotis et al. (2011) recovered as dominant fungal genera *Cladosporium* spp.,  
309 *Penicillium* spp. and yeasts. Due to the absence of an air filtration unit and the overlapping in the relative  
310 microbial air contamination, the authors ascribed the indoor presence of fungal contamination deriving

311 from the outdoor environment. The aerobiology of commercial dairy environments was investigated also  
312 by Soldatou, Psoni, Tzanetakis and Litopoulou-Tzanetaki (2006) through sedimentation technique and  
313 subsequent incubation. The authors isolated mainly micrococci and bacilli in two cheese factories making  
314 Feta cheese. Physiological and biochemical activities of abovementioned microflora were investigated  
315 too. The air contaminants exhibited acidifying and proteolytic activities potentially contributing to cheese  
316 ripening and flavor. More recently, Brandl et al. (2014) studied the bioaerosol in different sites of milk  
317 powder and powdered infant formula processing units of a dairy plant. As expected, due to the strict  
318 hygienic requirements of these environments, numbers of cultivable microorganisms were very low (<100  
319 CFU/m<sup>3</sup> of air) during production in filling, bagging and final packaging zones in comparison with other  
320 industrial locations. Additionally, following measurements on particle sizes of air, through handheld laser  
321 particle counters, the authors found a high correlation between total airborne particles in the size range 1  
322 to 5 µm and numbers of CFU. The authors concluded on the practical usefulness of a simple surveillance  
323 system based upon laser-mediated counting of airborne particles occurring in a specified size range.  
324 Simon and Duquenne (2014) referred on the airborne bioload, measured by an impactor sampler, in  
325 cheese-maturing cellars. Concentrations from 10<sup>3</sup> to 10<sup>6</sup> CFU/m<sup>3</sup> and from 10<sup>4</sup> to 2 × 10<sup>8</sup> CFU/m<sup>3</sup> were  
326 recorded for bacteria and fungi, respectively. Such levels resulted from 1 up to 5 log<sub>10</sub> cycles (brushing  
327 area) higher than those revealed in points of the plant considered uncontaminated. The authors concluded  
328 that throughout the process certain employees are exposed to high concentrations of airborne cultivable  
329 fungi.

330 Few studies focused on the composition of the microbiota present in the air of wineries, in particular  
331 on yeasts, both beneficial and spoilage ones (Ocón et al., 2013), and moulds (Ocón et al., 2011). An in-  
332 depth study on the microbial ecology in the air of a winery was recently reviewed by Pérez-Martín et al.  
333 (2014).

334 Overall, above discussed investigations remark the large variability of microbial air counts in food  
335 commercial plants as a function of a range of factors, including the sector, the hygienic requirements of  
336 each zone of the plant, the design, as well as processing conditions. To date, the legislator does not

337 impose any restriction on the number of airborne microorganisms being aware of the complexity of an  
338 ecosystem such as the air in the food industry. Nonetheless, the European Community Board (European  
339 Collaborative Action, 1996), in the context of the provision of healthy and environmentally sustainable  
340 buildings laid down a report on indoor air quality and its impact on man. In this document, the air of  
341 generic indoor environments (private houses, non-industrial workplaces and public buildings) was  
342 categorized in “very low” ( $< 50 \text{ CFU/m}^3$ ), “low” ( $50\text{--}100 \text{ CFU/m}^3$ ), “medium” ( $100\text{--}500 \text{ CFU/m}^3$ ) and  
343 “high” ( $> 500 \text{ CFU/m}^3$ ).

344

#### 345 **Air handling**

346 The food environment is often wet and includes many sources of aerosols contributing to microbial  
347 contamination, especially in critical areas where the products are exposed to air for long periods.  
348 Different physical mechanisms affect the movements of airborne particles resulting in a greater difficulty  
349 to control their movements. Generally, proper implementation of air-handling equipment can ensure that a  
350 large part of the airborne particles does not come into contact with exposed foods. An approach to reduce  
351 air microbial load consists in the filtration of air entering a specific area. Besides filtration, also a heating,  
352 ventilation and air conditioning (HVAC) system is widely used. This equipment allows the desired  
353 management of temperature and humidity of air as well as the flow direction and the pressurization within  
354 a specific area allowing to control airborne microorganisms. The latter are not inactivated, but possibly  
355 accumulated on the filter surface and can proliferate in case of high humidity ( $> 80 \%$ ). Generally, an air  
356 flow of 1.5 m/s or greater is required to ensure maintenance of one-way flow. Temperatures and relative  
357 humidity (likewise atmospheric gases, light, irradiation and surrounding organic material) are  
358 environmental factors associated with survival and growth of airborne microorganisms (Ijaz, Zargar,  
359 Wright, Rubino, & Sattar, 2016). Therefore, the control of these factors is desirable. To remove the heat  
360 load imposed by the processing environment (processes and people) and to provide employees with fresh  
361 air, 5–25 air changes per hour are considered sufficient. Proper ventilation removes also moisture released  
362 during processing and prevents condensation and the subsequent mold growth on surfaces. In addition, to



363 prevent bioaerosol contamination within HVAC systems, it is crucial to have a good understanding of the  
364 mechanisms of particle deposition and the subsequent fouling rate (Da et al., 2015). In food  
365 manufacturing facilities, the use of computational fluid dynamics programs is a useful tool for prediction  
366 of airflow movements inside specific areas. This approach supports the correct placement of air  
367 ventilation systems enhancing good sanitary of food processing environments (Skåra & Rosnes, 2016).

368

### 369 **Air disinfection**

370 In general, to inactivate environmental bioaerosols, different microbial decontamination technologies  
371 have been investigated. These include carbon nanotube filter, ion emissions, UV irradiation and  
372 electrostatic field (Liang et al., 2012). In the air of a food facility, type and amounts of microorganisms  
373 can vary widely as a function of the site and on a day-to-day basis in the same environment (Masotti et  
374 al., 2019). To strengthen preventive measures against air bioload, in view of attaining the goal of  
375 providing a safe and a high quality product to the consumer, food business operators are interested in the  
376 adoption of additional approaches other than regular sanitation procedures. In particular, chemical  
377 fogging, ozonation and UV irradiation of the air are major commercially available solutions. These  
378 techniques are currently implemented in the pharmaceutical and clinical sectors, but far from being  
379 common in food processing environments. Each of these techniques is characterized by benefits and  
380 drawbacks to be properly evaluated for effective disinfection (Table 2). In the food industry a steady  
381 growing interest is arising in these additional disinfection practices to minimize cross-contamination from  
382 the air, especially in critical areas (e.g., filling, packaging). One prerequisite for their effective  
383 implementation is the application to closed environments.

384

### 385 **Air disinfection by chemical fogging**

386 Fogging or aerosolization is the dispersion of a liquid in the form of fine mist in the air. Aerosolized  
387 disinfectants have been applied since many years for therapeutic use in the healthcare sector (Otter, Yezli,  
388 Perl, Barbut, & French, 2013). Subsequently, this technique has been implemented also in food factories

389 for decontamination of products (fruits and vegetables) (Oh, Gray, Dougherty, & Kang, 2005) or  
390 disinfection of surfaces in packaging or storage areas, process lines, cooling chambers (Holah, et al.,  
391 1995). Fogging is also used to reduce the counts of airborne viable microorganisms deriving from low-  
392 care areas, people, structures, or formed as aerosols during cleaning procedures (Burfoot, Hall, Brown, &  
393 Xu, 1999). The ultrafine droplet size of the dry fog prevents it from easily falling onto surfaces, a  
394 desirable quality for area decontaminations (Krishnan et al., 2012).

395 This technique has been also used quite widely by chilled food manufacturers, especially in high-  
396 care environments such as salad, sandwich, ready meal and dairy processing. Typically, the process  
397 requires at least 15–30 min for fog dispersion and proper chemical action. Subsequently, to allow settling  
398 of suspended droplets, a period of 45–60 min is necessary to reenter the treated room. Various types of  
399 delivery systems of the disinfectant solution in the air in the form of fine mist are available (Brown &  
400 Wray, 2014). Either a static purpose-built system with strategically placed nozzles or, more commonly, a  
401 mobile unit can be adopted (Holah, 2011). Over the years, fogging automatic systems developed. The  
402 engineering of devices, in particular the type of nozzle, is of primary importance for the success of the  
403 treatment. Checking nozzles for clogging and gaskets for integrity are preliminary steps to take before the  
404 disinfection treatment. Fogging is generally categorized, on the base of droplet size, into atomization (or  
405 nebulization) and aerosolization (Stanga, 2010). The former term is used when droplets have a diameter >  
406 30  $\mu\text{m}$ . These sizes result in shorter settling times, undesirable moistened surfaces and reduced  
407 disinfecting activity. Typically, with aerosolization, droplets of disinfectant are no wider than 5  $\mu\text{m}$ .  
408 Small sizes (within the range 0.5–5  $\mu\text{m}$ ) characterize droplets with non-wetting surface, longer suspension  
409 times and an electric charge as a consequence of friction during the aerosolization.

410 Fogging for air disinfection of food environments is a scarcely studied research topic (Bore &  
411 Langsrud, 2005). Burfoot et al. (1999) reported that in the chilled food industry, fogs were most effective  
412 when the diameter of droplets lied between 10 and 20  $\mu\text{m}$  giving a uniform coverage and a reasonable  
413 settling time (45 min). Up to 3  $\log_{10}$  cycle reduction was measured in air microbial counts as well as on  
414 upward-facing surfaces, by using an active concentration of 2 mg/mL of a quaternary ammonium

415 formulation. Smaller droplets allowed a good distribution, but the fog remained airborne for several  
416 hours, thus not allowing the entering of personnel in the working area. Bagge-Ravn, Gardshodn, Gram,  
417 and Fønnesbech Vogel (2003) in the slicing area of a salmon smokehouse evaluated the efficacy of  
418 peracetic acid-based fogging. After spread of a dense fog (mean droplet sizes of 15  $\mu\text{m}$ ) by a mobile unit,  
419 air was monitored by passive air sampling through settle plates exposed for 2 h in different spots of the  
420 room. The authors obtained a significant improvement of air hygiene level (expressed in terms of  
421 reduction of total aerobic counts). More recently, by test trials in dairy environments, Masotti et al. (2019)  
422 reported the effectiveness of hydrogen peroxide aerosolization in the inactivation of airborne  
423 microorganisms. The mist dispenser produced particles with diameters of 5–15  $\mu\text{m}$  of aerosolized  
424 hydrogen peroxide. Weekly-based air treatments in cheese making and packaging rooms lasted 16 and 20  
425 min, respectively, and were followed by 20 min of settling time to allow the aerosol decomposition.  
426 Following the post-treatment air sampling, microorganisms were almost absent during 5 weeks of  
427 investigation in the packaging room ( $< 10 \text{ CFU/m}^3$ ), whereas in the cheese making area only a slight  
428 number of bacteria (63  $\text{CFU/m}^3$ ) and molds (39  $\text{CFU/m}^3$ ) were enumerated. The occurrence of these  
429 residual molds (mainly represented by *Cladosporium herbarum*, *Penicillium* spp. and *Alternaria*  
430 *alternata*) was ascribed to recontamination from outdoor air and failures in the facility design.

431 Overall, major output from the literature on fogging disinfection outlined the facts that *i*) this  
432 technique should not be considered as a substitute of the regular cleaning and disinfection procedures; *ii*)  
433 further research is required to comprehensively evaluate the impact of parameters such as type of  
434 chemical, relative humidity and temperature; *iii*) the success of the aerosolization is related to the design  
435 of the treated area.

436

#### 437 **Air disinfection by ozone**

438 Ozone ( $\text{O}_3$ ) is a gas acting as a strong oxidizing agent and biocide (Marriott & Gravani, 2006). It has a  
439 broad-spectrum antimicrobial power, being active against bacteria, fungi, viruses, protozoa and bacterial  
440 and fungal spores (Pascual, Llorca, & Canut, 2007). For this reason, ozone has been used for decades for

441 water treatment. An extensive review on the principles of ozone treatment, the mechanism of action and  
442 applications in the food industry has been recently published (Brodowska, Nowak, & Śmigielski, 2017).  
443 In food processing environments the most advanced germicidal applications include food surface hygiene,  
444 sanitation of food plant equipment, treatment of food plant waste and reuse of waste water (Guzel-  
445 Seydim, Greene, & Seydim, 2004). Ozonation is performed after the cleaning step, because the  
446 germicidal activity is lost following its contact with residual organic material such as food debris. Several  
447 organizations and countries approved the use of ozone as antimicrobial agent for direct contact with  
448 drinking water and for food decontamination, including vegetables, fish, meat, poultry and dairy products  
449 (Brodowska et al., 2017; Christ, Savi, & Scussel, 2016; Tiwari & Rice, 2012). In recent years, ozonation  
450 has become more and more widely accepted as an eco-friendly “green” technology (O’Donnell, Tiwari,  
451 Cullen, & Rice, 2012). An increasing interest for ozone application resulted in the opinion of the Italian  
452 Ministry of Health (2010) endorsing the use of gaseous ozone for disinfecting empty cheese ripening and  
453 storage facilities. Portable ozone generators are now available. They have discharge units and fans to  
454 create the ozone at variable concentrations and catalytic converters to decompose ozone to oxygen after  
455 the treatment. Benefits related to the use of ozone consist in the easy access to hidden sites, being in the  
456 gaseous state. It has also the advantage of the absence of by-products, as it breaks down quickly into  
457 oxygen without leaving undesirable residues on either food or food contact surfaces. This technique  
458 allows both to save water in comparison to the use of other biocides and to improve the quality of  
459 wastewaters, for instance by avoiding the presence of harmful chlorine compounds. Furthermore, ozone is  
460 generated *in situ* on demand without the need to store it. On the other hand, some disadvantages consist in  
461 the high capital cost (i.e., the corona discharge generator). Despite this, ozone treatment remains more  
462 cost-effective than alternative treatment techniques.

463 Most studies focused on the effectiveness of ozone in the aqueous phase (ozonated water) against  
464 foodborne microorganisms attached to food contact surfaces or for food decontamination (Baumann,  
465 Martin, & Feng, 2009; Brodowska et al., 2017; Cullen & Norton, 2012). Only few published reports are  
466 available on the use of gaseous ozone. In this case, the disinfection treatment is carried out in confined

467 spaces, for long times (1–4 h vs 1–10 min of ozonated water; Pascual et al., 2007) generally overnight and  
468 in the absence of personnel. Ozone in the gaseous phase presents safety issues to humans, being a  
469 powerful irritant to the respiratory tract and a cellular poison that interferes with the ability of lungs to  
470 fight infectious agents (Marriott & Gravani, 2006). In the United States, the Occupational Safety and  
471 Health Administration (OSHA) recommends that ozone exposure must not be higher than 0.1 ppm by  
472 volume (the equivalent of 0.2 mg/m<sup>3</sup> of air) under normal working conditions for 8 h daily, or 40 h a  
473 week without adverse effects. Exposure to ozone at 0.1–1.0 ppm causes irritation to eyes, throat and nose  
474 as well as headaches. High levels (from 1.0 ppm up to 100 ppm) result in asthma-like symptoms (Pascual  
475 et al., 2007). Therefore, efficient systems for the detection and destruction of residual ozone after the air  
476 disinfection treatment speed up its decomposition and are reasonably required for the safety of  
477 employees. Foreseeing the potential risks, a continuous ozone analyzer, triggering a general alarm as soon  
478 as the concentration of ozone exceeds 0.1 ppm in the atmosphere of the ozonation room, should be  
479 installed. The above-mentioned term “safety” also refers to the equipment and instrumentation. Ozone  
480 may interact with the equipment and all surfaces. Therefore, it is essential to take into consideration only  
481 ozone-compatible materials.

482 In the dairy field, in particular in cheese ripening rooms, ozone gas proved to be effective in reducing  
483 the viable numbers of mold spores in the air. Serra, Abrunhosa, Kozakiewicz, Venâncio and Lima (2003)  
484 tested gaseous ozone treatments (overnight, during non-work time) for 20 weeks in a closed ripening  
485 room of unspecified cheese types. Ozone generated at a rate of 8 g/h for 12 h/d allowed obtaining a 10-  
486 fold reduction in the viable airborne mold loads to mean levels < 50 MPN/m<sup>3</sup> of air. Differently, the  
487 treatment did not affect the number of mold spores and hyphae on food contact surfaces, due to the short  
488 half-life of ozone. On this basis, according to the authors, gaseous ozone is useful to reduce the  
489 sedimentation of airborne molds on cheese surface during ripening. Pinto, Schmidt, Raimundo and  
490 Raihmer (2007), in the ripening room of extra-hard cheeses, carried out an environmental disinfection  
491 program consisting in the discontinuous generation of 0.48 mg of gaseous ozone per m<sup>3</sup> of air. Following  
492 a 40-day trial, the authors observed 1.5 log<sub>10</sub> reduction of fungal viable counts in the air, meanwhile a

493 lower but significant reduction was measured on cheeses surface ( $0.7 \log_{10}$  cycles). More recently,  
494 Masotti et al. (2019), investigated the effectiveness of air ozonation in the packaging room of a dairy  
495 factory over a 5-week period to reduce air contamination. The treatment realized overnight 3 h/d and for 3  
496 d per week meanly resulted in the absence of microbial growth in 92 % of air samplings, whereas the  
497 remaining ones were characterized for bioload levels  $< 20$  MPN/m<sup>3</sup>. The authors underlined the  
498 usefulness of a periodic air ozonation as a practical solution to counteract unexpected spike levels of  
499 bioaerosol due to uncontrolled factors.

500 In general, before installing an ozone generator, an *ad-hoc* tailored study is recommended to take into  
501 consideration factors specific to any processing environment. This approach can allow designing a safe  
502 and efficient program of air disinfection contributing to the implementation of food safety management.

503

#### 504 **Air disinfection by UV radiation**

505 Ultraviolet light in the frequency range 100–280 nm, categorized as UV-C, is an established means of  
506 disinfection. Radiation at short wavelengths (approximately 254 nm) allows inactivating microorganisms  
507 such as bacteria, viruses, protozoa, molds, yeasts and algae. This environmentally friendly technology is  
508 established to reduce microbial contamination in the public health field (hospitals, health care facilities,  
509 public shelters) and the pharmaceutical industry (Lee, 2011). In the food industry, UV-C irradiation is  
510 exploited to disinfect air, surfaces of plant, packaging materials, water as well as fruit and vegetables  
511 during post-harvest storage (Begum, Hocking, & Miskelly, 2009). The germicidal action mechanism  
512 consists in damaging deoxyribonucleic acid (DNA), thus rendering the microbes incapable of replicating  
513 (Kowalski, 2009). Microorganisms in the air are inactivated as a function of both the distance from the  
514 source of radiation and reflection. Lamps installed together with suitable coating materials (e.g., stainless  
515 steel and anodized aluminum) allow to reflect as much as 80% of the emitted radiation (Stanga, 2010).  
516 Currently, UV-C lamps used in air disinfection applications are low-pressure mercury vapor lamps.  
517 Innovation challenges consist in: *i*) lamp technology to develop more versatile and efficient lamps, *ii*) the  
518 use of nontoxic materials, in drivers and controls to adapt performance as a function of the need (e.g.

519 occupied/unoccupied room) and *iii*) systems to warn in case of malfunction (Miller, Linnes, & Luongo,  
520 2013). UV lamps prove to be very useful when coupled with high efficient air filters in air ducts and store  
521 rooms for seasoning, chilling and drying when foods cannot be removed (e.g., cheese, salami, Parma  
522 ham) (Stanga, 2010). UV energy is mainly applied after air passage through the HVAC air-handling  
523 ductwork (also called “in-duct” system) allowing an effective air microbial inactivation. Bacteria, viruses  
524 and molds that either grow or pass through the air handling system are reduced. In the real world of food  
525 environments, the irradiation at high intensities remains not accessible to personnel in the room. Lamp  
526 locations and air movement patterns within a room need to be considered for optimal disinfection. The  
527 inactivation of microorganisms is dependent on several parameters, including: *i*) the dose of radiation  
528 received (measured in  $J/m^2$ ), which is the product of intensity (measured in  $W/m^2$ ) and exposure duration  
529 (measured in s); *ii*) the wavelength of received radiation and *iii*) the microbial sensitivity to UV-C  
530 radiation (Reed, 2010). For instance, for 90 % inactivation of *Aspergillus niger*, *A. flavus* and *Penicillium*  
531 *roqueforti* the required UV-C doses are 132, 60 and 13  $J/m^2$ , respectively (Begum et al., 2009). This  
532 species-dependent response is a function of the composition of conidia, which can be either thin-walled  
533 and with light pigmentation or dark-pigmented due to melanin. The latter component is photo-protective  
534 and increases the survival and longevity of fungal spores, whereas non-melanin compounds are less  
535 defensive against UV-C radiation (Kowalski, 2009). The susceptibility of airborne microorganisms is also  
536 a function of temperature and relative humidity. There is a substantial lack of information on air-based  
537 UV constants. Furthermore, environmental conditions are known to affect UV light. For instance, as  
538 relative humidity increases, UV light becomes less efficient (Cutler & Zimmerman, 2011). The delivery  
539 of the required UV dose uniformly and consistently to large volumes of air is a significant challenge given  
540 the current state of the technology. To date the UV inactivation of bioaerosols is considered an added  
541 value in comparison to the standard chemical sanitation protocol alone.

542 Most research studies on UV irradiation are dedicated to food decontamination and water purification  
543 (Begum et al., 2009). Investigations on air as the target medium are scarce (Miller et al., 2013). Cundith,  
544 Kerth, Jones, McCaskey and Kuhlert (2002) reported that the use of wall-mounted germicidal air cleaning

545 units, using a combination of UV light and electrostatically polarized low-density media filter, proved to  
546 substantially reduce the risk of microbial contamination of meat products in a small meat processing  
547 plant. Under the conditions described by the authors, after 18 h of filtration a reduction from 1 to 1.5 log<sub>10</sub>  
548 in airborne bacteria and molds was observed. In bakeries, UV lamps are used on bread slicing equipment  
549 to minimize contamination from airborne molds (Begum et al., 2009). Recently, Yang, Zhang, Nunayon,  
550 Chan and Lai (2018) investigated the performance of UV irradiation through experiments evaluating  
551 exposure time, UV dose received and bacteria susceptibility. The authors confirmed that the ventilation  
552 duct UV germicidal irradiation system would potentially provide a supplementary solution for improving  
553 indoor air quality within mechanical ventilated/air-conditioned environments. Despite UV-C is an  
554 effective microbial inactivation means, a drawback limiting its application is the production of ozone, a  
555 molecule of concern for its healthy effects (Ryan, McCabe, Clements, Hernandez & Miller, 2010).

556

#### 557 **Air disinfection by cold plasma**

558 Air ionization is a decontamination technology primarily focused on liquids or surfaces (Arnold, Boothe,  
559 & Mitchell, 2004; Liang et al., 2012). Recently, this technique turned into the spotlight for the application  
560 in the food sector to reduce microbial contamination of food (Lacombe et al., 2015; Misra & Jo, 2017).  
561 Cold plasma has been recently investigated also for air sterilization (Liang et al., 2012; Zhou, Yang, Lai,  
562 & Huang, 2016). The principle of this technique consists in the passage of the air over an ionizing tube  
563 emitting high voltage discharge (in-duct system) resulting in positively and negatively charged ions,  
564 clusters of oxygen ions, oxygen-containing radicals, UV-C irradiation and a series of combined effects of  
565 these factors (Niemira, 2012; Zhou et al., 2016). These reactive chemical species attract naturally charged  
566 airborne micro-organisms, damaging their membranes, DNA and/or proteins. In addition, high-voltage  
567 electrical discharges result in the generation of ozone. Thus, monitoring schemes should be implemented  
568 to avoid the presence of excess ozone concentration in the treated room. Measures to remove the ozone  
569 should be evaluated if required. For the scale up to commercial treatment levels an optimization and a  
570 more complete understanding of these chemical processes is required. An additional aspect to take into



571 account for practical considerations is the cost of cold plasma tubes and the decrease in the emission of  
572 ion species with time (Lai, Cheung, Wong, & Li, 2016).

573 The in-duct cold plasma system is very useful for disinfecting large quantities of air as it passes  
574 through the HVAC system before its re-circulation. Obviously, this will only be useful for disinfection of  
575 contaminated air through the duct, but not at the sources, i.e. inanimate environmental surfaces (Lai et al.,  
576 2016). Despite recent appearance on market of cold plasma disinfection units for in-duct applications  
577 (Zhou et al., 2016), the limitation of this technology is the early stage of development and the variety and  
578 complexity of the necessary equipment.

579

## 580 **Conclusions**

581 In the course of time, the safety of food gained a high priority, because industry has been under pressure  
582 to deliver products minimally processed, more fresh in taste and appearance, with less preservatives and  
583 with prolonged shelf life. Thus, intervention strategies to control all vectors of food contamination should  
584 be pursued. Bioaerosols in a food facility may be potential contributors to food spoilage. Due to factory  
585 air movements, a complete environmental control is complex and almost impossible. In the design of new  
586 factories, proper planning in locating air inlets, extracts, doorways and processing equipment is of utmost  
587 importance to optimize air movements. The periodic monitoring of microbial levels in the air is useful to  
588 identify potential sources of contamination. Intervention should be taken to maintain a bioaerosol load  
589 consistent with the hygienic requirements of the food product. Through years, air disinfection techniques  
590 such as chemical aerosolization, ozonation and UV irradiation evolved providing a feasible and cost-  
591 effective solution for the decontamination of selected areas of the facility. Air decontamination can entail  
592 the benefit of reducing microbial settling on frequently touched or food contact surfaces, thus preventing  
593 the risk of microbial spread. Furthermore, the implementation of a proactive approach based on scheduled  
594 air disinfection treatments would be an ancillary strategy, especially in case of inadequate hygiene of  
595 structures.

596

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**Figure caption**

Figure 1 – Overview of major sources/vectors of microbial contamination and their interactions in the food industry.

ACCEPTED MANUSCRIPT

Table 1 – Pros and cons of air sampling techniques available for the food industry.

Sampler	Air sampling	Pros	Cons	Use in real food industry
Settle plate	Passive	Easy and cheap device to monitor generic air bioload. No cell stress by reduced viability.	Qualitative method, based on collection by “fall out”. Biased to larger particles. Sensitive to air movements.	++
Impactor	Active	Multiple choice of devices (slit and sieves). Practical in industrial use. Information on size distribution. Used to recover viruses.	Cost of device.	++
Cyclone separator	Active	Available as portable hand-held instrument. Practical in industrial use. Less cell stressing than impaction methods.	Selective for large air particles. Tendency to higher counts than other air samplers.	++
Filter	Active	Not expensive. Simple to operate. Suitable for enumeration of moulds and bacterial spores. Used also to recover viruses.	Possible stress by cell desiccation.	+
Impinger	Active	Useful for heavily contaminated air environments.	Impractical in industrial use. Sterilization of the device after each use. Possible loss of survivability.	+
Electrostatic precipitator	Active	Useful for collection of viruses or sensitive microbial strains. Compatible with analysis by polymerase chain reaction.	No literature in food sector.	–

++, frequent use; +, occasional use; –, not used.

Sources: H. M. L. Lelieveld, M. A. Mostert, & J. Holah, 2005. *Handbook of hygiene control in the food industry*. Oxford, UK: Woodhead Publishing Limited; Ljungqvist & Reinmüller, 2007; Verreault, Moineau, & Duchaine, 2011; Reponen, 2017.

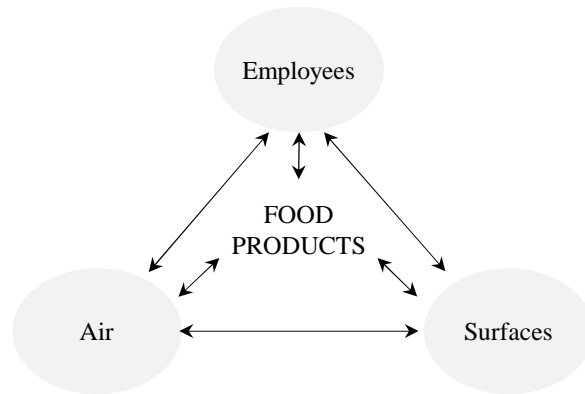
Table 2 – Pros and cons of disinfection techniques available in food industry for air treatment.

Disinfection technique	Pros	Cons	Use in food industry
Air filtration and UV irradiation	Disinfection efficacy of in-duct UV-C lamps.	Energy consumption. Increase of temperature of air supply. Fungi can escape UV radiation.	++
Chemical aerosolization	Wide spectrum of efficacy against microorganisms. Environmental friendliness (as a function of the agent used). Dry aerosol.	Time for aerosolization and chemical action. Sealing of treated environments. Controlled room re-entry, to avoid safety issues. Equipment material compatibility.	+
Ozone gas	Excellent antimicrobial activity. Production <i>in situ</i> . Immediate action. Auto-decomposition. Lack of residues on food.	Health and safety issues in case of uncontrolled room re-entry. Need of a gaseous ozone analyzer. Absence of personnel and food. Use of sealed environments. Corrosive to several soft metals and rubber. Cost of ozone generator.	+
UV irradiation	Discrete disinfection efficacy. No use of chemicals. Synergistic effectiveness when in tandem with other technologies (e.g., photocatalysis, air filtration).	Health effects due to the production of ozone as a by-product. Delivery of sufficient UV irradiation to large volumes of air. Influence of environmental conditions.	+
Cold plasma	Disinfection efficacy in air duct flow. Static purpose-built system or mobile unit.	Health effects due to the production of ozone as a by-product. Cost of cold plasma tubes. No up-scale for commercial applications. Lack of research data on air disinfection effectiveness in food environments.	–

++, frequent use; +, occasional use; –, not used.

Sources: Burfoot, Hall, Brown, & Xu, 1999; Marriott & Gravani, 2006; Pascual, Llorca, & Canut, 2007; Stanga, 2010; Krishnan et al., 2012; Cutler & Zimmerman, 2011; O'Donnell, Tiwari, Cullen, & Rice, 2012; Christ, Savi, & Scussel, 2016; Zhou, Yang, Lai, & Huang, 2016; Yang, Zhang, Nunayon, Chan, & Lai, 2018; Chen et al., 2019; Masotti et al., 2019.

Figure 1



**Highlights**

- Indoor air is a vector of contamination in the food industry
- Sampling and analysis of bioaerosol is a strategy to prevent food contamination
- Air disinfection is a task of interest in the real world of food environment
- Ozonation, UV irradiation, chemical fogging are feasible air disinfection techniques