

Effects of disinfectants on inactivation of mold spores relevant to the food industry: a review

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1 2	Effects of disinfectants on inactivation of mold spores relevant to the food industry: a review.
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20 Abstract

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Due to the dissemination of airborne conidia and spores, molds can contaminate various 22 surfaces. In the food industry sector, their presence and development can have health and 23 economic implications. In order to control these undesirable microorganisms, various 24 approaches can be used but the main one relies on the use of disinfectants. The objective of 25 this review is to report the existing studies on the effect of various disinfectant molecules (i.e., 26 sodium hypochlorite, chlorine dioxide, ethanol and other alcohols, hydrogen peroxide, 27 peracetic acid, and quaternary ammonium compounds) on the inactivation of fungal spores. 28 These studies were sorted depending on the targeted fungal species. Noteworthy, in the food 29 30 industry, four log and three log reductions are required to claim a fungicidal activity for suspension (European Standard 1650, 2019) and surface (European Standard 13697/IN1, 31 2019) treatments, respectively. Most of the presented studies concerned Penicillium and 32 Aspergillus species (44 and 31% of the literature, respectively). In general, for a given 33 disinfection procedure, ascospores were more resistant than conidia, and Aspergillus conidia 34 35 were more resistant than Penicillium ones. However, the variability of encountered molds (e.g. species, strains, physiological state) and disinfection procedures (e.g. molecules, 36 concentrations, contact time) affected the efficacy of disinfectants. 37

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41 **1. Introduction**

Quality is a major issue for food industries as various hazards can affect food 42 products. Among microbiological hazards, fungal contamination is a major spoilage source of 43 many food products such as fruits, vegetables, bakery and dairy products. Mold development 44 leads not only to modification of organoleptic properties but can also have safety implications 45 in the case of a mycotoxigenic species. Consequently, food spoilage by fungi is synonymous 46 47 of economic losses for producers, and food waste and health risk for the consumer (Bryden, 2007; Coton and Dantigny, 2019; Dagnas and Membré, 2013; Sengun et al., 2008). As for the 48 49 medical sector, pathogenic strains of *Candida* and *Aspergillus* species are the primary cause of hospital-acquired fungal infections (Seeliger and Schröter, 1984), and in recent years these 50 infections have been recognized as serious threats to immunocompromised patients (Bodey et 51 al., 1992; Morrison et al., 1994; Wildfeuer et al., 1998). Moreover, other mold-associated 52 53 infections have constantly increased over the last decades (e.g. mucormycosis, Morin-Sardin et al., 2017). 54

In order to prevent microbial contamination, food manufacturers mainly use disinfectants. Due to the presence of at least one active ingredient with antimicrobial activity, disinfectants are microbiocidal products. Antimicrobial activity is defined as the ability of a biocide to reduce the number of viable cells. More generally, disinfectants have a broad activity range, such as a bactericidal, virucidal, sporicidal, yeasticidal and fungicidal activity.

Fungicidal activity is determined by means of the log reduction of the number of viable mold spores. This is a key factor in the choice of a disinfectant. This activity is evaluated according to specific standards drawn up by international or national agencies, and specifying standardized test methods (van Klingeren *et al.*, 1998). In Europe, standards for assessing fungicidal activity have been established by the European Committee for Standardization (CEN/TC 216). These standards indicate various experimental factors such as

the species and strains, the culturing and harvesting methods, the test procedure (contact time,
concentrations, temperature...), the enumeration method as well as the criteria for assessing
the antimicrobial activity of the tested biocide.

The fungicidal effect of disinfectants is based on the inactivation of reference 69 microorganisms, Candida albicans (ATCC 10231) and Aspergillus brasiliensis (ATCC 70 16404), either by the suspension test, the carrier test or the airborne room disinfection 71 procedure (Table 1). The European Standard 1275 (2006) entitled "Quantitative suspension 72 test for the evaluation of the basic fungicidal activity of antiseptics and chemical 73 disinfectants" consists in demonstrating a basic activity of the test product in optimal 74 75 conditions of use. A suspension test is described to establish whether the test product has a fungicidal activity based on reducing by at least 4 log the reference microorganisms. The 76 European Standard 1650 (2019) entitled "Quantitative suspension test for the evaluation of 77 78 the fungicidal or yeasticidal activity of antiseptics and chemical disinfectants used in the food industry, in industry, in domestic and community areas", assesses the activity of the 79 disinfectant under practical conditions of use. A suspension is tested in the presence of an 80 interfering substance simulating clean or dirty conditions. The European Standard EN 81 13697/+A1 (2019) entitled "Chemical antiseptics and disinfectants - Quantitative test of a 82 non-porous surface for the evaluation of the bactericidal and/or fungicidal activity of 83 chemical disinfectants used in the food industry, in industry, in domestic and community 84 sectors. Test method without mechanical action and requirements" describes a test on a non-85 porous surface intended to assess the efficacy of the disinfectant molecule diluted in hard 86 water. The ability of the latter to reduce, by at least 3 log, the reference microorganisms after 87 15 min or less exposure, at a temperature between 18°C and 20°C is also evaluated in the 88 presence of an interfering substance simulating clean or dirty conditions. 89

The surface test is by far the most important, challenging and representative of the 90 91 tests of disinfectant efficacy. With the exception of drinking water, the surface test is more relevant than the suspension test because it is truer to practical conditions and theoretically, 92 microorganisms attached to a surface will be more resistant than those in a suspension, 93 therefore this presents the greatest challenge. The quantitative surface test evaluates test 94 suspensions of bacteria and fungi in a solution of interfering substances, designed to simulate 95 96 clean and dirty conditions, which are inoculated onto a test surface and dried (Sandle, 2017). More recently, the European Standard 17272 (2020), described a test to disinfect by an 97 automated process the surfaces of the overall area including the external surfaces of the 98 99 equipment contained in rooms. This standard aims at simulating practical conditions of airborne disinfection in a laboratory situation. 100

101 There are several sanitizing chemical compounds approved for use on food contact 102 surfaces, such as hypochlorites, chlorine dioxide, iodophors, peroxides, ethanol, quaternary ammonium. Regardless of the chemical, the stock solution must be diluted to a specific 103 104 concentration to ensure maximum efficacy. The list of authorized compounds may vary with 105 time and according to countries. Final concentrations in use for some sanitizing compounds approved in the Canada and in the US are listed Table 2. Some compounds, such as ethanol, 106 107 do not require any rinsing. For the other compounds, the maximum residue levels that remain in the food product should be evaluated as acceptable by the safety authorities in Canada. In 108 Europe, disinfectants (biocides) used in the food industry are controlled by a range of 109 legislation, but two are key in determining the level of disinfectant that can be taken up by 110 foodstuffs after the disinfectant's legitimate use. Regulation (EC) No 396/2005 (2005) on 111 maximum residue levels of pesticides in or on food and feed of plant and animal origin 112 governs the use of pesticide residues. Regulation (EU) No 528/2012 (2012), concerning the 113

making available on the market and use of biocidal products, governs the use of biocides forthe purpose of disinfection in the food hygiene sector.

Despite the importance of fungal contamination in the food industry, in the medical 116 environment, and in domestic and community areas, to the best of our knowledge, the effect 117 of disinfectants on molds spores was not reviewed. With the notable exception of Aspergillus 118 *fumigatus*, and *Trichophyton mentagrophytes*, this review was concerned with mold species 119 relevant to the food industry. A. fumigatus is the most common species implicated in all 120 pulmonary syndromes and some contaminated food could be a potential source of exposition 121 in immunosuppressed patients (Bouakline et al., 2000). In Canada, T. mentagrophytes is the 122 123 reference organism for assessing the fungicidal efficacy of a disinfectant in the medical area (Gaulin et al., 2011). In this context, this review reported the effects of various biocides 124 against the inactivation of more than sixty different mold species. This work synthesized for 125 126 the main disinfectants used, their chemical nature, mode of action, efficacy on mold spores through studies published over the past twenty years. 127

In the context of this review, disinfectant efficacy was usually assessed by means of the log reduction in viable spores, $log(N/N_0)$, where N was the number of survivors after treatment, and N₀ the initial load of spores which, according to the European standards, should be in the range 1.5-5 10^7 CFU/ml. When the N₀ value was not stated in a study, the effect of the disinfectant could not be evaluated and the publication was not used, as for example Roberts and Reymond's (1994) study.

In many studies, disinfection treatments were applied to fruits, vegetables, or grains artificially contaminated by sprayed spore suspensions. The efficacy of treatments were not assessed by enumerating the number of survivors but by determining a decay number (Karabulut et al., 2004, 2005) or a contamination percentage (Andrews, 1996). None of these responses could be correlated to the number of survivors after treatment. Accordingly, this

kind of studies was also discarded. In water applications, the disinfectant effectiveness was often expressed in terms of the Ct (concentration x time_{reaction}) value needed to inactivate a certain percentage of the population (Pereira et al., 2013). Therefore, in all studies reported in Table 3, the impact of the treatments was always expressed in terms of log reduction. When no survivors were detected after treatment, the log reduction was indicated as greater than log N. Most studies assessed the fungicide effect of disinfectants for a given exposure duration, but studies examining inactivation kinetics were scarce.

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147 **2.** Chlorine compounds

148 2. 1. Generalities

Chlorine compounds are a family of biocides consisting of a chlorine atom responsible 149 for oxidative activity. The main disinfectants in this chemical group are hypochlorous acid, 150 151 hypochlorite ion, chlorine and chlorine dioxide. Hypochlorous acid, hypochlorite ion, and chlorine are referred to as free (Fukuzaki, 2006; Leidholdt, 2000) or active (EU regulation 152 N°528/2012, 2017) chlorine. Formation of the latter compounds results from the 153 decomposition of sodium hypochlorite in water, 1% w/w sodium hypochlorite being 154 equivalent to 0.96% w/w active chlorine (EU regulation N°528/2012, 2017). Other chlorine 155 156 sources correspond, for example, to sodium dichloroisocyanuric acid or calcium hypochlorite after dissolution in water. The first solution of sodium hypochlorite was produced by the 157 French chemist Claude Berthollet in the 18th century under the name "Eau de Javel" and its 158 antimicrobial properties were discovered in the 19th century by the French chemist Antoine 159 Germain Labarraque (Mupparapu and Kothari, 2019). Chlorine compounds are widely used in 160 the food industry to disinfect surfaces or in hospitals to disinfect material, but also in the 161 treatment of drinking water and some ready to eat food such as salads (Beuchat and Ryu, 162 1997; Gómez-López et al., 2009; Jeffrey, 1995; Okull et al., 2006; Rossman et al., 1994). The 163

main factor that influences the antimicrobial activity of chlorine compounds is the presence of 164 165 soiling/organic load which will have a negative impact by reacting with chlorine (Boothe, 1998; Kotula et al., 1997). Neutral pH has a positive impact on antifungal activity of sodium 166 hypochlorite solution by promoting the formation of hypochlorous acid (pKa 7.54) which is 167 more active than the hypochlorite ion predominant in alkaline solution (pH 9-11) (Boothe, 168 169 1998; Leidholdt, 2000; McDonnell and Russell, 1999). Chlorine concentrations are usually 170 expressed in ppm (mg/l) for disinfecting water and in percentage for sanitization of food industry surfaces. 171

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2. 2. Mode of action

Antimicrobial activity of a sodium hypochlorite solution is due to the oxidative action 174 of hypochlorous acid and hypochlorite ion on cellular components. These compounds are able 175 176 to react with multiple biomolecules such as lipids, amino acids, proteins, peptides and nucleic acids. Therefore, chlorine compounds are able to target many cellular structures and 177 compounds such as cell wall, cell and mitochondrial membrane, enzymes as well as DNA 178 (Fukuzaki, 2006). Available chlorine inhibits the metabolism by oxidation of thiol functions 179 or oxidation and chloramination of amino groups present in the membrane and cytoplasmic 180 enzymes (Denyer and Stewart, 1998). In addition to disrupting the metabolism, hypochlorous 181 acid has been shown to induce the production of Reactive Oxygen Species (ROS) with a 182 lethal effect on cells (Dukan et al., 1999). Sodium hypochlorite and chlorine dioxide have also 183 been shown to exert a genotoxic effect as observed on the Saccharomyces cerevisiae yeast 184 (Buschini, 2004). Electron microscopic examination showed that A. fumigatus conidia 185 exposed to sodium hypochlorite solution were smaller and smoother than the untreated 186 187 conidia, with loss of surface structures and some surface padding compared to untreated conidia (Martyny et al., 2005). 188

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2. 3. Antifungal activity of sodium hypochlorite

Bundgaard-Nielsen and Nielsen (1996) tested the fungicidal efficacy of 3% sodium 191 hypochlorite on twenty-five fungal contaminants. After 10 minutes exposure, inactivation 192 values ranged from 1.7 to more than 5.2 log. Twenty-one isolates exhibited more than 4 log 193 inactivation. Ascospores, except those of *Eurotium repens* (=*Aspergillus pseudoglaucus*) 194 195 showed a greater resistance than conidia to sodium hypochlorite. For example, *Monascus* ruber and Neosartorya pseudofischeri (=Aspergillus thermomutatus) ascospores showed 196 inactivation of 2.9 and 4.0 log, respectively. In contrast to these ascospores, all conidia, with 197 198 the exception of one *Penicillium commune* isolate (1.7 log inactivation), exhibited more than 4 log reduction. However, the fungicidal activity of sodium hypochlorite can vary within the 199 same species. At a concentration of 0.3 % for a 10 min exposure, the inactivation of 200 201 *Penicillium roqueforti* ranged from 0.5 to 4.5 log depending on the considered strain (n=2).

A low sodium hypochlorite concentration at relatively short exposure times was sufficient to ensure effective inactivation of *Aspergillus niger* and *P. roqueforti*, both widely distributed contaminants in the food industry. Indeed, three minutes exposure to 0.5% sodium hypochlorite solution were sufficient to completely inactivate (about 6 log inactivation) all *A. niger* isolates (n=5) and less than 1 minute was sufficient for all tested *P. roqueforti* isolates (n=5) (Korukluoglu et al., 2006).

A lower pH, and the presence of certain non-ionic surfactants, may increase the antifungal activity of sodium hypochlorite. Okull et al. (2006) showed that a *Penicillium expansum* suspension exposed for 5 min to sodium hypochlorite at 50 ppm, induced 1.9 log and 0.5 log decrease in viable conidia at pH 6.3 ± 0.2 and pH 8.5 ± 0.2 , respectively. At the lower pH, the addition of nonionic surfactant such as Span 20, Tween 20 or Tween 80 at 0.01% (v/v) led to the absence of viable spores. At the higher pH, only Span 20 and Tween 20
at 0.1% gave a significant reduction of 2.4 log.

Furthermore, it has been observed that, at concentrations lower than 0.1%, the 215 fungicidal activity of sodium hypochlorite decreases sharply. The contact times required for 216 the complete inactivation of *Penicillium brevicompactum* conidia at sodium hypochlorite 217 concentrations of 0.053% was 30 min as compared to 4 and 2 min for 0.299 and 0.525 % 218 concentrations, respectively (Ebling, 2008). A contact time of 15 min to 1% chlorine allowed 219 220 total inactivation of Aspergillus ochraceus (Gupta et al., 2002). The fungicidal efficacy of sodium hypochlorite on Aspergillus flavus, A. fumigatus and A. niger isolates was further 221 evaluated by Mattei et al. (2013). The recommended concentration of 0.4% sodium 222 hypochlorite for 72h was sufficient to inactivate one third of the Aspergilli (n=18). Another 223 third required 0.8 and 1.6%, while the remaining third exhibited survivors for 1.6%. These 224 225 authors concluded that sodium hypochlorite was the least effective disinfectant against Aspergillus spp. as compared to chlorhexidine-cetrimide, benzalkonium chloride and a 226 227 chlorophenol derivative. It should be underlined that 72h exposure time exceeds the maximum time that can be allowed in the food industries, even for a complete disinfection. 228

Frison et al. (2015) showed that a decrease in pH from 9 to 6.3 reduced the contact 229 time required to achieve 4 log inactivation for five isolates of Aspergillus section Nigri. For a 230 700 mg/l (0.07%) sodium hypochlorite solution, according to the considered strain, the 231 contact time required to reach 4 log at pH 9 and at pH 6.3, ranged from 16.2 to 28.8 min, and 232 from 9.5 to 14.7 min, respectively. However, at higher sodium hypochlorite concentrations, 233 the effect of pH on the contact time required to reach 4 log inactivation was less noticeable. 234 For 1300 mg/l (0.13%) sodium hypochlorite, the contact time associated with 4 log 235 inactivation ranged from 1.1 to 1.2 min at pH 6.3 and 1.4 to 2 min at pH 9. 236

Pereira et al. (2013) investigated the impact of pH and temperature on the efficacy of 237 238 two free chlorine concentrations (1 and 3 mg/l) to inactivate A. fumigatus, Aspergillus terreus, Cladosporium cladosporioides, Cladosporium tenuissimum, Penicillium citrinum, Penicillium 239 griseofulvum and Phoma glomerata, molds which are found in different source waters. 240 Efficacy was evaluated by means of an inactivation rate constant expressed in min⁻¹, the 241 greater the inactivation rate, the more efficient the inactivation was. At 21°C, a pH decrease 242 from 7 to 6 increased the inactivation rate about 8-fold for *P. glomerata*. This was explained 243 by the higher proportion of hypochlorous acid (HOCl), a more powerful disinfectant, than 244 hypochloride ion (OCl⁻) at pH 6. An increase from 1 mg/l to 3 mg/l increased also the 245 246 inactivation rates about 3 fold for Aspergilli, C. cladosporioides, and P. citrinum. A lower impact was observed for C. tenuissimum, P. griseofulvum and P. glomerata. Eventually, at 1 247 mg/l, the impact of the type of water on inactivation was highlighted. Inactivation rates were 248 249 about 2-8 fold greater in laboratory grade water than in settled water (i.e. obtained for decantation of treated wastewater). At pH 7, a temperature decrease from 21°C to 4°C led to a 250 251 decrease in inactivation rate of about 2-4 fold only depending on the considered species. In 252 contrast to this study, less than one log reduction difference was observed between A. brasiliensis conidia submitted to sodium hypochlorite (0.5 and 1% for 5-15 min) at 10 and 253 40°C (Stefanello et al., 2021). 254

Drinking water contaminated with *A. fumigatus, Aspergillus versicolor* and *Penicillium purpurogenum* was also tested using sodium hypochlorite and monochloramine solutions (Ma and Bibby, 2017). Disinfectant concentrations and contact times were in the range 1-4 mg/l, and 0-60 min respectively. The Ct values needed to obtain 3 log inactivation ranged from 48.99 to 194.7 mg.min.l⁻¹ and from 90.33 to 531.3 mg.min.l⁻¹, for sodium hypochlorite and monochloramine, respectively. *A. versicolor* and *P. purpurogenum* isolated 261 from a drinking water system were more resistant than an *A. fumigatus* clinical isolate to both262 disinfectants.

The efficacy of sodium hypochlorite solution (0.1%, 0.5%) and 1% of active chlorine) 263 on A. brasiliensis as well as on fungal food spoilers, namely C. cladosporioides, P. commune, 264 Penicillium polonicum and P. roqueforti, was tested by Bernardi et al. (2018) according to the 265 EN 13697 standard (2001). Mold conidia were fixed on stainless steel coupons in the 266 267 presence of an interfering solution, then sodium hypochlorite solutions were applied for 15 minutes. Under these conditions, sodium hypochlorite was the most effective disinfectant 268 against all tested species compared to the other disinfectants tested in the study (i.e. 269 270 benzalkonium chloride, biguanide, peracetic acid, and quaternary ammonia). Active chlorine 0.1% was very effective against C. cladosporioides with more than 4 log inactivation and 271 effective against A. brasiliensis with 3 log inactivation. However, the same concentration 272 273 showed insufficient fungicidal activity against the three tested Penicillium species (2 log reduction). On the contrary, 0.5% active chlorine was very effective against all species with 274 275 more than 4 log inactivation. The greatest sodium hypochlorite concentration tested in this 276 study, 1%, was above the concentrations usually allowed for use in food industries.

Potential intraspecific response heterogeneity to sodium hypochlorite exposure was 277 278 evaluated on several strains of Aspergillus pseudoglaucus (n=2), Hyphopichia burtonii (n=3), Penicillium paneum (n=3) and P. roqueforti (n=3), (Bernardi et al., 2019). Exposure to 0.2% 279 active chlorine for 15 min had very limited effect against all tested strains with a mean 280 inactivation value of 1.37 log for A. pseudoglaucus and 2.30 log for P. paneum. At 0.2% 281 active chlorine, the greatest difference in inactivation within the same species was observed 282 for P. roqueforti with a 0.54 log inactivation difference between the most resistant strain (1.74 283 284 log) and the most sensitive one (2.28 log). However, amongst all the concentrations tested (0.01%, 0.1% and 0.2%), the greatest difference in inactivation was obtained at 0.1% active 285

chlorine on *P. paneum* with a 1.03 log difference between the most resistant strain (0.04 log 286 287 reduction) and the most sensitive strain (1.07 log reduction). Although sodium hypochlorite is used in a relatively wide concentration range from 0.01 to 2% depending on the target mold, 288 cleanliness conditions and desired level of hygiene, active chlorine concentrations lower than 289 or equal to 0.2% do not guarantee an inactivation greater than 3 log on treated surfaces. 290 Sodium hypochlorite and biguanide were the least effective, reducing less than 3 log from 291 292 initial control, not being the most suitable agents for the control of toxigenic fungi (Aspergilli) 293 in food industries (Gonçalves Lemos et al., 2020).

On the other hand, disinfectant treatments of three *Penicillium* species isolated from fruit juices were evaluated according to the EN 13697 standard (2015) on stainless steel, in the absence or presence of sucrose as interfering substance (Nierop Groot et al., 2019). Remarkably, for *Penicillium buchwaldii* conidia, chlorine from hypochlorite inactivation was enhanced in the presence of sucrose (1.8 log reduction with sucrose compared to 0.9 log in the absence without).

300 Evaluation of sodium hypochlorite fungicidal activity has been extended to other types 301 of material than the traditional steel coupon. The spores of Alternaria alternata, A. niger, Cladosporium herbarum, Mucor bainieri, Penicillium chrysogenum, Stachybotrys chartarum 302 and T. mentagrophytes were inactivated by 2.4% sodium hypochlorite to undetectable levels 303 in glazed and unglazed ceramic carriers. Test organisms were non-culturable in 10/10 trials, 304 after 5-min contact times in glazed ceramic carriers, and after 10 min in unglazed ceramic 305 carriers, representing a>3-log10 to>5-log10 reduction, depending on the mold type (Reynolds 306 307 et al., 2012).

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2. 4. Antifungal activity of chlorine dioxide

In their study, Bundgaard-Nielsen and Nielsen (1996) found that 5% chlorine dioxide had a fungicidal efficacy similar to 3% sodium hypochlorite on their selection of target fungi (see §2.3). For 10 minutes exposure, inactivation values ranged from 2.7 log to more than 5.2 log. Eleven out of 20 isolates exhibited more than 4 log inactivation. All isolates exhibited more than 3 log reduction except *Penicillium caseifulvum* (2.7 log).

The impact of pH, temperature and humic acid concentration on the fungicidal activity 315 316 of chlorine dioxide (ClO₂) in the range 0.5-3.0 mg/l was assessed on *Cladosporium* sp., Penicillium sp., and Trichoderma sp. (Wen et al., 2017). The inactivation rate constants for 317 chlorine dioxide, 2 mg/l, were 5, 26 and 17 fold those obtained for chlorine, 2 mg/l, for 318 Cladosporium sp., Penicillium sp., and Trichoderma sp., respectively. Inactivation rate 319 constants were not significantly affected by pH 6 and 7. In contrast to pH, inactivation rate 320 constants decreased significantly by lowering temperature from 27 to 10°C (by 25% for 321 322 Penicillium sp. and 50% for Cladoporium sp. and Trichoderma sp.). While the inactivation rate constants did not vary significantly in the 0-2 mg/l humic acid range, inactivation rates at 323 324 0.4 mg/l were half those obtained at 0.2 mg/l. Overall, by increasing susceptibility order, the species were ranked *Penicillium*<*Trichoderma*<*Cladosporium*. 325

As shown in the study of Nierop Groot et al. (2019), chlorine dioxide remained effective against *Penicillium* spp. at low temperatures. At 7°C, for 5 min contact time to 500 ppm chlorine dioxide an inactivation greater than 5 log was shown for *Penicillium bialowiezense*, *P. buchwaldii* and *P. expansum*. In comparison, the inactivation obtained by chlorine was 2.5 log in the case of *P. bialowiezense* and less than 1 log for the two other species. In another study (Okull et al. 2006), *P. expansum* exposed to chlorine dioxide at 3 ppm for 5 min exposure exhibited 3.7 log inactivation.

Regarding the effect of chlorine dioxide on ascospore-producing species, 10 min exposure to 5% chlorine dioxide led to more than 4.5, more than 4 and 3.3 log inactivation for

E. repens (=A. pseudoglaucus), M. ruber and N. pseudofischeri (=A. thermomutatus), 335 336 respectively (Bundgaard-Nielsen and Nielsen, 1996). Dijksterhuis et al., (2018) also evaluated the fungicidal effect of this biocide on heat-resistant ascospores of Aspergillus fischeri, 337 338 Paecilomyces variotii, Paecilomyces niveus and Talaromyces macrosporus (=Penicillium macrosporum) which were either dormant or activated by 80°C heat treatment for 5 min. For 339 the first three mentioned species, the two kind of ascospores were inactivated after 500 and 340 1000 ppm treatments for 5 min. Only T. macrosporus (=P. macrosporum) ascospores 341 survived up to 500 ppm, but full eradication of this species (i.e. inactivation \geq 5 log) was 342 observed after 30 min. During viability assessment experiments, heat activated ascospores of 343 344 this species showed larger colonies than dormant ascospores, thus suggesting an increased resistance of heat activated ascospores to the disinfectant. 345

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2. 5. Antifungal activity of sodium dichloroisocyanuric acid

In their 1996 study, Bundgaard-Nielsen and Nielsen also tested the fungicidal efficacy 348 349 of sodium dichloroisocyanuric acid at 0.37 g/l. After 10 minutes exposure, inactivation values 350 ranged from 1.3 log to more than 5.2 log according to the considered target fungi (see §2.3), seven isolates exhibiting more than 4 log inactivation. Eight strains were particularly resistant, 351 namely two P. commune isolates (1.7 and 3.7 log reduction), P. caseifulvum (1.9 log 352 353 reduction), Penicillium discolor (1.9 log reduction), Penicillium nalgiovense (1.7 log reduction), P. roqueforti (0.9 log reduction) and Penicillium carneum (1.7 log reduction) as 354 well as *E. repens* (= *A. pseudoglaucus*) ascospores (0.3 log reduction). 355

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357 3. Alcohols

358 *3. 1. Generalities*

Alcohols used as disinfectants correspond to ethyl alcohol (or ethanol), isopropyl 359 360 alcohol (or isopropanol) and n-propanol. However, ethanol is the most commonly used molecule in this family. Alcohols are water soluble molecules consisting of a polar hydroxyl 361 362 function and a hydrocarbon tail. Alcohols have long been used for their antimicrobial activity and the first evaluations of their efficacy can be traced back to 1881 with the work of Robert 363 Kock (Price, 1939). In the food industry, the use of alcohols as disinfectant has the advantage 364 of being safe for the user (food-grade) and of not leaving any residue on the treated surface 365 due to their rapid evaporation. Alcohols can be sprayed, fumigated or applied through 366 impregnated wipes. Alcohols, like ethanol, are formulated in relatively strong concentrations. 367 368 Their antimicrobial activity is optimal in the range 60-90% (Boyce, 2018a; Jeffrey, 1995; McDonnell and Russell, 1999). 369

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3. 2. Mode of action

Alcohols act on the cell membrane and cytoplasmic proteins. These molecule toxicity 372 373 depends on their aliphatic chain and hydrophobicity. This is why small chain alcohols, like 374 ethanol, are toxic at high concentrations. The mode of action of ethanol on fungal cells was reviewed by Dao and Dantigny (2011). Ethanol molecules will interact with cell membrane 375 lipids at the lipid-water interface leading to increased polarity and decreased cell permeability 376 to polar compounds. Cell death at high ethanol concentrations is believed to be associated 377 with an increase of the fluidity and then the disruption of their cell membrane followed by 378 leakage of cellular compounds (Ingram and Buttke, 1985; Jones and Greenfield, 1987; 379 380 Tesnière, 2019, Russell, 2003). Ethanol has several deleterious effects on the integrity of the cell membrane. At high concentrations, ethanol dissolves membrane lipids and denature 381 382 proteins on the membrane leading to destruction of the cell membrane (Bacílková, 2006; Ingram and Buttke, 1985). However, it seems that thinning of the membranes also takes place 383

at low concentrations of ethanol. At higher ethanol concentrations (> 20%), ethanol would 384 385 transform the phospholipid bilayer into an interdigitated phospholipid phase (Henderson and Block, 2014; Simon and McIntosh, 1984). In the interdigitated phospholipid phase, the 386 387 membrane lipids of the phospholipid bilayer are distributed over a single layer leaving the terminal methyl group of the acyl chains exposed. This configuration is a consequence of the 388 interactions between the hydroxyl group of the short alcohol chains (C \leq 3) with the polar 389 390 heads of the phospholipids and their non-polar half with the acyl chains of the phospholipids. Since the nonpolar part of these alcohols is smaller than the acyl part of phospholipids, empty 391 spaces are created in the inner part of the membrane. These spaces will be filled by lipids 392 393 from the opposite side leading to the formation of an interdigital phase (Simon and McIntosh, 1984; Weber and de Bont, 1996). Interdigital phase membranes have a lower thickness and 394 greater permeability than the natural state of the membrane. 395

396 The modification of the biophysical properties of membranes also disrupts the function of membrane proteins. In addition, alcohols can interact directly with proteins and 397 denature them. Due to their ability to diffuse across the plasma membrane, alcohols are 398 responsible for the denaturation of cytoplasmic proteins. Protein denaturation is believed to be 399 an important factor in the fungicidal activity of alcohols (Boyce, 2018). Given the role of 400 proteins in the proper functioning of cells, their denaturation affects the metabolism and 401 absorption of nutrients in the cell (Ingram and Buttke, 1985). Moreover, ethanol has been 402 reported to damage mitochondrial membranes (Dao and Dantigny, 2011) and DNA (Ibeas and 403 Jimenez, 1997). Mitochondria are thought to have a close role in ethanol-induced death in S. 404 405 cerevisiae (Carmona-Gutierrez et al., 2012).

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3. 3. Antifungal activity of ethanol

Bundgaard-Nielsen and Nielsen (1996) tested the fungicidal efficacy of 70% ethanol 408 409 on target species (see §2.3.). For 10 minutes exposure, inactivation values ranged from 1.3 to more than 5.2 log and eleven isolates exhibited more than 4 log inactivation. M. ruber 410 411 ascospores were the most resistant spores (1.3 log reduction) thus suggesting that ascospores had a greater resistance than conidia to ethanol. The age of ascospores seems to have an 412 influence on the fungicidal activity of ethanol as 110-day M. ruber ascospores were more 413 414 resistant to 70% ethanol than 21-day ascospores (3.5 log difference). An important difference 415 in inactivation of approximately 4 log was observed between the most sensitive isolate and the most resistant *P. roqueforti* isolate, indicating intraspecific susceptibility heterogeneity. 416

417 Concentration and contact time are important factors that explain the antifungal activity of ethanol. However, other factors can affect the efficacy of ethanol. Dao et al. (2008) 418 showed that temperature, water activity and the mode of application impacted P. 419 420 chrysogenum, Penicillium digitatum and Penicillium italicum inactivation by ethanol. In this study, P. digitatum was the most sensitive to ethanol, while P. chrysogenum was the most 421 422 resistant mold. Increasing the ethanol preparation water activity from 0.7 to 0.9 a_w resulted in 423 a greater inactivation of P. chrysogenum. For this species, ethanol solutions were more effective than ethanol vapors. For the more sensitive species, namely P. digitatum and P. 424 *italicum*, vapor ethanol was more effective at 30°C than at 10°C. The experimental condition 425 "0.7 a_w , 30°C, liquid application for 4 days, 10% w/w ethanol" was the most drastic tested 426 condition and ensured total inactivation of the three tested species with inactivation values 427 ranging from 3.19 log to 5.78 log. 428

Biological factors of the target mold had also a great effect on the fungicidal activity of ethanol. Dao and Dantigny, (2009) tested the effect of ethanol vapors at 0.30 and 0.45 kPa on hydrated and dry-harvested conidia of *P. chrysogenum*, *P. digitatum* and *P. italicum*. After 432 24 hours exposure, all dry-harvested conidia remained viable, while the hydrated conidia had

mean inactivation values of approximately 1, 3.5 and 2.5 log for P. chrysogenum, P. 433 434 digitatum and P. italicum, respectively. The fungicidal efficacy of ethanol vapors on dryharvested conidia of P. chrysogenum, P. digitatum and P. italicum was further evaluated by 435 Dao et al. (2010). For a 24-hour exposure, 0.7 kPa ethanol vapor pressure inactivated 4 log P. 436 digitatum and P. italicum conidia, but only 3.2 log P. chrysogenum conidia. While an ethanol 437 vapor pressure of 1.5 kPa achieved an inactivation of about 4.5 log and more than 5 log for *P*. 438 chrysogenum and the two other species, respectively. Fungicidal activity of ethanol was also 439 impacted by the water activity of the spore production medium (Dao and Dantigny, 2009). 440 Conidia produced from mycelium grown at 0.85 a_w were more resistant to 0.67 kPa ethanol 441 vapors than conidia produced at 0.99 a_w . After 48 h exposure, 6.5, 5.1 and 5.9 log inactivation 442 were obtained for conidia produced at 0.99 a_w for P. chrysogenum, P. digitatum and P. 443 *italicum*, respectively. Conidia produced at reduced a_w (0.90-0.95) required 120 h contact 444 445 time to reach 4.5 log inactivation for P. chrysogenum and P. digitatum, a 6 log reduction was observed at 98 h and 120 h exposure for P. *italicum* conidia produced at 0.95 and 0.90 a_w , 446 447 respectively. The greatest resistance of dry-harvested conidia produced at a reduced water activity was also observed on four isolates of P. commune exposed to a 70% ethanol (Visconti 448 et al., 2020). For 1 min exposure, regardless of the isolates, only dry-harvested conidia 449 produced at a_w 0.950 exhibited survivors. Survival after 2 min exposure (and even 3 min) for 450 this physiological state was only observed for P. commune UBOCC-A-116003. For this 451 strain, the impact of the physiological state was greater than 1.54 log reduction between dry-452 harvested conidia produced at 0.950 a_w , that exhibited survivors after 1 min treatment, and the 453 3 other kinds of conidia that were all inactivated. 454

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3. 4. Antifungal activity of isopropanol

According to Bundgaard-Nielsen and Nielsen (1996), 70% isopropanol was less effective than 70% ethanol to ensure the total inactivation of the great majority of the target species (see §2.3.). After 10 minutes exposure, inactivation values ranged from 0.9 to more than 5.2 log, and eleven isolates exhibited inactivation values greater than or equal to 4 log. *E. repens* (=*A. pseudoglaucus*) and *M. ruber* ascospores, but also *P. roqueforti* conidia, were more resistant to 70% isopropanol than the other selected isolates with inactivation values of 2.3, 0.9 and 3.5 log, respectively.

Korukluoglu et al. (2006) reported a significant intraspecific variability in the resistance of *P. roqueforti* conidia to an isopropanol-based disinfectant. While, three isolates were completely inactivated (approximately 6 log reduction) after 2 to 8 minute treatments, two others could survive for more than 60 min exposure. In contrast to *P. roqueforti*, lower intraspecific heterogeneity was noticed for *A. niger*, 6 log reduction being obtained for treatments ranging from 10 to 25 min depending on the considered isolates.

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471 **4. Peroxides**

472 *4. 1. Generalities*

Peroxides are chemical species characterized by the presence of two covalently bond 473 oxygen atoms (R-O-O-R'). In disinfection, the most widely used peroxides are hydrogen 474 475 peroxide (H₂O₂), peracetic acid (CH₃COOOH) and ozone (O₃). These disinfectants, which are powerful oxidizers, can be used in liquid or gaseous form by misting, nebulization or 476 vaporization. Hydrogen peroxide and peracetic acid are both sensitive to soils, such as organic 477 matter, and more effective at temperatures greater than 25°C. Unlike peracetic acid, hydrogen 478 peroxide is more effective at alkaline pH albeit more unstable (Stanga, 2010). Due to its lower 479 480 sensitivity to organic load and peroxidases, peracetic acid is considered more effective than hydrogen peroxide (McDonnell and Russell, 1999). Peracetic acid has a limited impact on the 481

environment and human health due to its decomposition into water and acetic acid (Alvaro et
al., 2009; Lee and Huang, 2019; Taverner et al., 2018), thus making peracetic acid an
alternative of interest to chlorine compounds that can form chloramines.

485

486 *4. 2. Mode of action*

Hydrogen peroxide and peracetic acid produce ROS which are powerful oxidizing agents of proteins by acting on their thiol groups (Denyer, 1995). Hydrogen peroxide can act on other cellular components such as ribosomes, cell and mitochondrial membranes as well as DNA (McDonnell and Russell, 1999; Qin et al., 2011; Russell, 2003). In *P. expansum*, Qin et al., (2011) showed that the accumulation of ROS following exposure to hydrogen peroxide was responsible for oxidative damage on the proteins of the mitochondrial membrane leading to the collapse of the membrane potential.

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4. 3. Antifungal activity of hydrogen peroxide

Bundgaard-Nielsen and Nielsen (1996) reported that 10 min exposure to 3% hydrogen 496 peroxide had almost no effect on *P. roqueforti* conidia and *E. repens* (=*A. pseudoglaucus*) 497 ascospores (0.3 and 0 log reduction, respectively). As shown by Martin and Maris (2012), 498 Absidia corymbifera, A. versicolor, Geotrichum candidum, Penicillium cyclopium and 499 Scopulariopsis brevicaulis exhibited 3 log inactivation at 12.5, 1.71, 0.50, 2.34, 5.51% for 15 500 min exposure. Formic, propionic, acetic, oxalic, lactic and sulfuric acids had synergistic 501 activity with hydrogen peroxide. In most cases (92%), the addition of these organic acids 502 503 reduced the effective fungicide concentrations by more than 4 fold. Among these six acids, only three retained this synergy when tested in the presence of an interfering agent. They were 504 505 corresponded to formic acid, against A. corymbifera, G. candidum and S. brevicaulis,

propionic acid against *A. corymbifera*, and *A. versicolor* and acetic acid against *A. corymbifera*.

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4. 4. Antifungal activity of peracetic acid

The effect of 0.3% peracetic acid was the same as 3% hydrogen peroxide on P. 510 roqueforti and E. repens (=A. pseudoglaucus) (Bundgaard-Nielsen and Nielsen, 1995). In 511 contrast to these authors who reported 0.3 log inactivation in P. roqueforti, 5 to 16 min 512 exposure to 0.3% peracetic acid inactivated 6 log of the latter species (Korukluoglu et al., 513 2006). The same authors indicated a stronger resistance of A. niger than P. roqueforti to 514 peracetic acid as more than 55 min were necessary to obtain a 6 log reduction in A. niger 515 conidia. According to Frison et al. (2015), 6.2-13.8 min exposure to 0,6% peracetic acid 516 517 decreased viable A. niger conidia by 4 log.

518 The effect of paracetic acid was also evaluated on conidia inoculated on stainless steel coupons (Bernardi et al, 2018). A 15 min treatment by 0.15% peracetic acid was not sufficient 519 520 to inactivate 4 log P. roqueforti conidia. These authors showed that 1.5% peracetic acid 521 succeeded in inactivating more than 4 log of C. cladosporioides, P. commune, P. polonicum and P. roqueforti conidia but failed to inactivate 3 log of A. brasiliensis conidia. The same 522 authors extended their study on coupons to other molds i.e., A. pseudoglaucus, P. paneum and 523 P. roqueforti, and disinfectants (benzalkonium chloride, biguanide and quaternary 524 ammonium, all tested at 0.3, 2.5 and, 5%) (Bernardi et al., (2019). At 3%, peracetic acid was 525 more effective than the other disinfectants at their highest concentrations to inactivate all the 526 tested species. P. paneum and one P. roqueforti were the most sensitive species to 3% 527 peracetic acid, exhibiting more than 4 log reduction, while A. pseudoglaucus was the most 528 529 resistant. Eventually, it was shown that A. brasiliensis proved less resistant than ascosporeforming molds tested, i.e., Chaetomium globosum, Talaromyces bacillisporus (=Penicillium 530

bacillisporum) and Aspergillus hiratsukae (Scaramuzza et al. 2020). In contrast to 531 benzalkonium chloride, a clearer higher antifungal efficacy of peracetic acid at higher 532 temperatures was demonstrated (Stefanello et al., 2021). When studying the efficacy of 533 534 electrolyzed water and sanitizers against heat resistant molds, Stefanello et al. (2020) found out that peracetic acid at the maximum concentration of 1% was the best antifungal agent 535 against Paecilomyces variotii, Paecilomyces niveus, Paecilomyces fulvus, Paecilomyces sp. 536 and Aspergillus neoglaber. Peracetic acid showed the best inactivation of Aspergillus from 537 Circumdati section spores (Gonçalves Lemos et al., 2020). 538

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540 **5. Quaternary ammonium compounds**

541 *5. 1. Generalities*

Quaternary ammonium compounds (QACs) are cationic surfactants. The cationic part 542 543 consists of a central positively charged nitrogen atom covalently bond with 4 alkyl or aryl groups. The cationic part is most often linked to a chlorine or a bromide to form a QAC salt. 544 545 Among the most widely used QACs, one can mention benzalkonium chloride, twin-chained 546 QAC dimethyl dodecyl ammonium chloride or bromide (DDAC or DDAB, 12), cetyltrimethylammonium bromide (CTAB) and cetylpyridinium chloride (CPC) (Jennings et 547 al., 2015). QACs are usually formulated with nonionic detergents to increase detergency 548 activity. The presence of a high concentration of calcium and magnesium ions in water, 549 soiling matter (Jeffrey, 1995) or lipids in organic matter (Boothe, 1998) has a negative impact 550 on QAC efficacy. (Jeffrey, 1995). QACs are not compatible with soaps or ordinary anionic 551 detergents; if the latter are used for cleaning, they must be rinsed off before applying QACs 552 otherwise the disinfectants will see its efficacy largely impacted. QACs are more effective in 553 554 alkaline conditions than in acid ones and a rise in temperature increases their activity. At the

usage dilutions, QACs are usually non-corrosive to surfaces but strong concentrations can
corrode mild steel or iron (Jeffrey, 1995).

557

558 *5. 2. Mode of action*

QACs are membrane disruptors, membrane disorganization would be due to electrostatic interactions between the QACs and the cell membrane lipids (potentially the phospholipids) at the intramembrane region level. This leads to the leakage of low molecular weight content (potassium ions, inorganic phosphates, amino acids, etc.), destruction of proteins and nucleic acid as well as lysis of the cell wall following the action of autolytic enzymes (Jennings et al., 2015; McDonnell and Russell, 1999; Russell, 2003).

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5. 3. Quaternary ammonium compounds antifungal activity

567 In their extensive disinfectant study, Bundgaard-Nielsen and Nielsen (1996) tested the fungicidal efficacy of four QAC-based disinfectants on the same previously mentioned 23 568 569 isolates (see § 2.3). For a 1.5% benzalkonium chloride-based disinfectant, after 10 minutes 570 exposure, inactivation ranged from 1 to more than 5.2 log according to the considered isolate. Fourteen isolates exhibited inactivation greater than or equal to 4 log, namely E. repens (=A. 571 pseudoglaucus) (> 4.5 log), M. ruber (> 4.1 log) and N. pseudofischeri (=A. thermomutatus) 572 573 (> 4.5 log) ascospores. One out of two A. versicolor, P. discolor, P. roqueforti and P. roqueforti were characterized by log reduction greater than 4 log. Exposure to a 2% N-alkyl-574 dimethyl-ammonium chloride-based disinfectant for 10 minutes led to inactivation values 575 ranging from 2.1 to more than 4.2 log (for S. brevicaulis). Overall Penicillia were less 576 resistant than Aspergilli. Exposure to 2% dodecyl- + dodecyl-benzyldimethyl-ammonium 577 578 chloride-based disinfectant (same exposure time), inactivated from 1.5 log (A. versicolor) to more than 5.2 log spores (A. flavus, A. niger and Penicillium crustosum). The latter species 579

were also resistant to 10 minutes exposure to a mixture of benzalkonium chloride,
formaldehyde, glutaraldehyde and 2% cetalkonium chloride-based disinfectant.

Korukluoglu et al., (2006) tested the sensitivity of five A. niger isolates and five P. 582 roqueforti isolates to three commercial QAC disinfectants based on alkyl-dimethyl-583 benzylammonium chloride (product A, 2%), on didecyl-dimethyl-ammonium chloride 584 (product B, 0.5 and 1%), and on benzalkanium chloride (product C, 0.5, 1 and 1.5%). Product 585 586 B was the less efficient, at 1%, 8-21 min and more than 45 min were necessary to completely inactivate P. roqueforti and A. niger, respectively. Product A and product C, at 1%, 587 completely inactivated P. roqueforti and A. niger in maximum contact times of 3 and 5 min, 588 589 respectively. Benzalkonium chloride concentrations required to inactivate 2.7 log conidia were 0.25-2% for A. flavus, less than 0.125-0.5% for A. fumigatus and 0.25% for A. niger 590 (Mattei et al., 2013). 591

592 Benzalkonium chloride 2% demonstrated a poor efficacy (< 2 log reduction) when applied to A. brasiliensis, C. cladosporioides, P. commune, P. polonicum and P. roqueforti 593 for 15 min (Bernardi et al., 2018). However, a maximum efficacy, more than 4 log reduction, 594 was achieved for C. cladosporioides by increasing the concentration up to 3.5%, but a 595 concentration of 5% failed to reach this maximum efficacy for all the other species. This 596 observation was latter extended to A. pseudoglaucus (Bernardi et al., 2019). Another un-597 specified quaternary ammonium at 5% reached this maximum efficacy for C. cladosporioides, 598 P. commune and P. polonicum (Bernardi et al., 2018), but not for A. pseudoglaucus, P. 599 paneum and some P. roqueforti isolates (Bernardi et al., 2019). Benzalkonium chloride and 600 601 iodine were the most effective sanitizers to eliminate Aspergillus from the Flavi and Nigri section (Gonçalves Lemos et al., 2020). 602

603

604 **6.** Conclusions

In the agri-food-industry context, cleaning and sanitation are crucial steps to ensure 605 606 both microbial safety (absence of pathogens) and quality (absence of spoilers) in the final product. As for the sanitation aspect, there is a wide variety of active substances that can be 607 608 used and that are characterized by different chemical structure, mode of action, physicochemical properties and interactions with the environment. Moreover, each active 609 substance can be found in a variety of commercial disinfectants that differ in their 610 formulation. An effective disinfectant should be broad spectrum (i.e. eliminating Gram-611 positive and -negative bacteria as well as yeast and molds), act quickly, active even in the 612 presence of organic load, non-aggressive on the treated surface, safe for the environment, the 613 614 operator, the consumer and should not form by-products (Stanga, 2010). On one hand, the efficacy of a disinfectant can be improved by modifying the physico-chemical properties of 615 the disinfectant solution or by adding co-formulants that are not always disclosed by 616 617 manufacturers (Okull et al., 2006; Dao, 2008; Pereira et al., 2013; Wen et al., 2017). On the other hand, presence of soiling substances significantly decreases the efficacy of disinfectants. 618 619 The effectiveness of disinfectants generally increases at higher concentrations. Stefanello et 620 al., (2020) reported that the lowest concentration specified in various sanitizers label was ineffective in most cases (94%), thus suggesting that the greatest concentrations should be 621 preferred. However, increasing disinfectant concentrations is synonymous for a greater 622 consumption of product at an increased cost and leads to more time and water are necessary to 623 rinse the disinfectant. Then, some biocides, such as chlorine compounds, release by-products 624 (e.g. chloramines) that can have impacts on the consumer health and the environment. In this 625 626 context, although efficient, potentially deleterious substances should be substituted for more health and environmentally friendly products, provided that these disinfectants are fungicidal 627 628 enough.

Although A. brasiliensis (ATCC 10231) is the standard strain for assessing antifungal 629 630 efficiency of sanitizers, the European Standards suggested to extend these assessments to other species. This review suggested not to use C. cladosporioides as an additional species 631 632 because this species was found to be very sensitive to many disinfectants. Disinfectants should be tested against microorganisms present in each industry and veterinary/medicine 633 places, and selected according to their ability to inactivate the most resistant species. The 634 635 choice of the sanitizer is not easy, because disinfectant molecules perform differently not only depending on the considered species but also on the considered isolate within the same 636 species; therefore, intraspecific resistance heterogeneity should also be taken into 637 638 consideration. Real contaminations are due to highly hydrophobic airborne conidia. For this reason, liquid sanitizers contain non-ionic surfactant to speed-up the action of the fungicidal 639 molecules. However, airborne conidia remain dry, whereas conidia used in standard assays 640 641 are wet in spore suspensions. As demonstrated, hydration has a significant effect on the physiological state of fungal spores and their susceptibility to disinfectant, therefore this 642 643 aspect should also be taken into consideration. Overall, the choice of a multi-target efficient 644 disinfectant is not an easy task as numerous abiotic and biotic factors can impact its efficacy.

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Table 1: European standards for fungicidal assessment of disinfectants

European	Year	Areas	Method	Interfering	Experimental	Required
Standard				substances	conditions	log
						reduction
EN 1275	2006	Not stated	Suspension	none	20°C/15 min	≥4
			test			
EN 1650	2019	Food, industrial,	Suspension	Bovin serum	4-40°C/1-60	≥4
		domestic and	test	albumin / skimmed	min	
		institutional		milk		
EN 13624	2019	Medical	Suspension	Bovin serum	4-30°C/5-60	≥4
			test	albumin / sheep	min	
				erythrocytes		
EN	2019	Food, industrial,	Carrier test	Bovin serum	4-40°C/1-60	≥3
13697/+A1		domestic and		albumin / skimmed	min	
		institutional		milk		
EN 17272	2020	Medical,	Airborne room	Bovin serum	20°C/≤ 15h	≥4
		veterinary, food,	disinfection	albumin / skimmed	/50-75% RH	
		industrial, domestic	by automated	milk / erythrocytes/		
		and institutional	process	yeast extract		

Table 2: Concentrations of some sanitizers approved in the Canada and the US for use on food contact surfaces.

Compounds	Utilisation	Recommended	Reference	Country
	range	concentrations		
Hypochlorites	50-500 ppm	200 ppm (non porous)	Gaulin et al. 2011	Canada
		800 ppm (porous)		
		200 ppm	Rasin, 2021	US
Chlorine dioxide		100 ppm	Rasin, 2021	US
Iodophors	6.5-75 ppm		Gaulin et al. 2011	Canada
		25 ppm	Rasin, 2021	US
Peracetic acid	50-350 ppm	150-200 ppm	Gaulin et al. 2011	Canada
		100-200 ppm	Rasin, 2021	US
Hydrogen	Powder in 3 and		Gaulin et al. 2011	Canada
peroxide	6%			
		80-600 ppm	Rasin, 2021	US
Quaternary	200-1000 ppm	200 ppm	Gaulin et al. 2011	Canada
ammonium	100-400 ppm	200 ppm	Rasin, 2021	US

Organism	Disinfectant	Concentration	Application conditions	Fungicidal effect	Reference
Absidia corymbifera	Hydrogen peroxide	Up to 50% (m/v)	Liquid / 15 min 20°C	3 log (12.5%)	Martin and Maris, 2012
Alternaria alternata	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	3.5-3.6 log (5 min)	Reynolds et al., 2012
Aspergillus australensis	Benzalkonium chloride	0.3-2%	Surface / 15 min	2.0-4.9 log	Stefanello et al., 2020
	Biguanide	2-5%	Surface / 15 min	1.0-2.9 log	
	Iodine	0.2-1%	Surface / 15 min	1->5 log	
	Peracetic acid	0.3-1%	Surface / 15 min	3-3.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	2-3.9 log	
	Ortho-phenylphenol	15% 1g/m ³	Fumigation / 7h	2-2.9 log	
Aspergillus aureoluteus	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-2.9 log	Stefanello et al., 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Iodine	0.2-1%	Surface / 15 min	2-4.9 log	
	Peracetic acid	0.3-1%	Surface / 15 min	1-1.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-1.9 log	
	Ortho-phenylphenol	15% 1g/m ³	Fumigation / 7h	3-3.9 log	
Aspergillus brasiliensis	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1-3.9 log	Bernardi <i>et al.</i> , 2018 Stefanello <i>et al.</i> 2021
	Biguanide hexamethylene	2-5%	Liquid / 15 min	<1 log	Bernardi <i>et al.</i> , 2018
	Peracetic acid	0.15-3% 0.3-1%	Liquid 15 min Liquid 5 – 15 min	1-3.9 log 0.94-5.67 log	Bernardi <i>et al.</i> , 2018 Stefanello <i>et al.</i> 2021
	Quaternary ammonium	0.3-5%	Liquid 15 min	<1-2.9 log	Bernardi et al. 2018
	Sodium hypochlorite	0.1-1% 0.5-1%	Liquid 15 min Liquid 5 -15 min	3->4 log 1.10-2.67 log	Bernardi <i>et al.</i> 2018 Stefanello <i>et al.</i> 2021
	Iodine	0.2-1%	Liquid $5 - 15$ min	1.17-3.50 log	Stefanello <i>et al.</i> 2021
	Peracetic acid	0.1%	Surface / 40°C	4 log (20-60 s)	Scaramuzza <i>et al</i> . 2020

Table 3: Fungicidal effect of disinfectants on different mold species, concentration used and application conditions.

Aspergillus carbonarius	Peracetic acid	0.3-1%	Surface / 15 min	2->5 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	1-2.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-3.9 log	
	Iodine	0.2-1%	Surface / 15 min	1- 4.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-1.9 log	
Aspergillus fischeri	Chlorine dioxide	50-1000 ppm	Liquid / 5-60 min	>5 log (500 ppm/5 min)	Dijksterhuis et al., 2018
Aspergillus flavus	Sodium hypochlorite	0.2-0.8%	Surface / 2-8 min	<4.2 log (0.2-0.8%/2 min and 0.4%/ 2-8 min)	Andrews, 1996
	Benzalkonium chloride	0.0125-0.4%	Liquid / 72h	>2.7 log (0.0125-0.2%)	Mattei et al., 2013
	Sodium hypochlorite	0.1-1.6%		>2.7 log (0.4-1.6%)	
	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2->5.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>5.2 log	
	Ethanol	70%	Liquid / 10 min 20°C	4 log	
	Isopropanol	70%	Liquid / 10 min 20°C	4 log	
	Sodium dichloroisocyanuric acid	0.37 g.L ⁻¹	Liquid / 10 min 20°C	4.3 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	4 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	4.3 log	
	Peracetic acid	0.3-1%	Surface / 15 min	1-2.9 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	<1-1.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-4.9 log	
	Iodine	0.2-1%	Surface / 15 min	1->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-1.9 log	
Aspergillus fumigatus	Benzalconium chloride	0.0125-0.4%	Liquid / 72h	>2.7 log (0.0125-0.05%)	Mattei et al., 2013

	Chlorophenol derivative	0.0175-0.6%		>2.7 log (0.075-0.3%)	
	Sodium hypochlorite	0.1-1.6%		>2.7 log (0.4-0.8%)	
	Free chlorine	48.99-194.7	Liquid	4 log	Ma and Bibby, 2017
		mg.min.L ⁻¹	рН 7; 22.5°С	(60.43-77.33 mg.min.l ⁻¹)	
	Monochloramine	90.33-531.3	Liquid	4 log	
		mg.min.L ⁻¹	pH 8; 22.5°C	(120-136.7 mg.min.l ⁻¹)	
	Sodium hypochlorite	$1-3 \text{ mg.L}^{-1}$ free	Liquid	$2 \log (946 \text{ mg.min.l}^{-1})$	Pereira et al., 2013
		chlorine	pH 7; 25°C		
Aspergillus	Peracetic acid	0.1%	Surface / 40°C	4 log (175-300 s)	Scaramuzza et al. 2020
hiratsukae					
Aspergillus niger	Benzalconium chloride	0.0125-0.4%		>2.7 log (0.025%)	Mattei et al.,2013
	Chlorophenol derivative	0.0175-0.6%		>2.7 log (0.15-0.30%)	
	Sodium hypochlorite	0.1-1.6%		>2.7 log (0.4%)	
	Sodium hypochlorite	100-1300 mg.l ⁻¹	Liquid / 1-30 min	4 log (700-1300 mg.l ⁻	Frison <i>et al.</i> , 2015a
			pH 6.3 ; 9, 20°C	¹ /1.1-28.8 min)	
	Peracetic acid	750-6000 mg.l ⁻¹			
	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	4.1-5.3 log (5 min)	Reynolds et al., 2012
	Isopropyl alcohol	Un-diluted	Liquid / 1-60 min	6 log (10-25 min)	Korukluoglu <i>et al.</i> , 2006
	Peracetic acid	0.1, 0.3%	Liquid / 1-60 min	$6 \log (0.3\% / \ge 55 \min)$	
	Iodine	0.5, 1%	Liquid / 1-60 min	6 log (1% / 12-49 min)	
	Formaldehyde	0.2;0.5%	Liquid / 1-60 min	6 log (2-15 min)	
	Quaternary ammonium compounds	0.5-2%	Liquid / 1-60 min	6 log (1% / <2->51 min)	
	Chlorine	0.5-2%	Liquid / 1-60 min	6 log (0.5% / <1-3 min)	
	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2.7->5.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>5.2 log	
	Ethanol	70%	Liquid / 10 min 20°C	>5.2 log	
	Isopropanol	70%	Liquid / 10 min	>5.2 log	

			20°C		
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	2 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>5.2 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>5.2 log	
	Peracetic acid	0.3-1%	Surface / 15 min	1-1.9 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-3.9 log	
	Iodine	0.2-1%	Surface / 15 min	1-3.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-2.9 log	
Aspergillus nomius	Peracetic acid	0.3-1%	Surface / 15 min	1-1.9 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-3.9 log	
	Iodine	0.2-1%	Surface / 15 min	1-4.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-2.9 log	
Aspergillus ochraceus	Chlorine	1%	Liquid / 0.25-24h	>6.5 log	Gupta et al., 2002
	Peracetic acid	0.3-1%	Surface / 15 min	>5 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-3.9 log	
	Iodine	0.2-1%	Surface / 15 min	2->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-2.9 log	
Aspergillus parasiticus	Sodium hypochlorite	0.2-0.8%	Surface / 2-8 min	<4.1 (0.2-0.8%/2 min and 0.4%/ 2-8 min)	Andrews, 1996
	Peracetic acid	0.3-1%	Surface / 15 min	>1-1.9 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	>1-1.9 log	

	Benzalkonium chloride	0.3-2%	Surface / 15 min	1->5 log	
	Iodine	0.2-1%	Surface / 15 min	<1-3.9 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	<1-1.9 log	
Aspergillus	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1.10-1.56 log	Bernardi et al., 2019
pseudoglaucus			Surface / 15 min / 20°C	0.1-1.9 log	Bernardi et al., 2021
	Biguanide	2-5%	Liquid / 15 min	1.01-2.02 log	Bernardi et al., 2019
		0.3-5%	Surface / 15 min / 20°C	1-1.9 log	Bernardi et al., 2021
	Peracetic acid	0.15-3%	Liquid / 15 min	0-3.36 log	Bernardi et al., 2019
			Surface / 15 min / 20°C	1-2.9 log	Bernardi et al., 2021
	Quaternary ammonium	0.3-5%	Liquid / 15 min	1.12-1.79 log	Bernardi et al., 2019
	Sodium hypochlorite	0.01-0.2%	Liquid / 15 min	0-1.50 log	Bernardi et al., 2019
		0.01-1%	Surface / 15 min / 20°C	0.9-1.9 log	Bernardi et al., 2021
	Ortho-phenylphenol	1g/m^3	Fumigation	0 log	Bernardi et al., 2021
Aspergillus terreus	Sodium hypochlorite	1-3 mg.l ⁻¹ free	Liquid	$2 \log (1404 \text{ mg.min.l}^{-1})$	Pereira et al., 2013
		chlorine	рН 7; 25°С		
Aspergillus	Free chlorine	48.99-194.7	Liquid	$4 \log (112.1 \text{ mg.min.l}^{-1})$	Ma and Bibby, 2017
versicolor		mg.min.l ⁻¹	рН 7; 22.5°С		-
	Monochloramine	90.33-531.3	Liquid	4 log (707.8 mg.min.l ⁻¹)	
		mg.min.l ⁻¹	рН 8; 22.5°С		
	Quaternary ammonium	1.5, 2%	Liquid / 10 min	1.2-4 log	Bundgaard and
			20°C		Nielsen, 1995
	Quaternary ammonium	2%	Liquid / 10 min	2-3 log	
	+ aldehydes		20°C		
	Ethanol	70%	Liquid / 10 min	3.3->5.2 log	
			20°C		
	Isopropanol	70%	Liquid / 10 min	3.3->5.2 log	
			20°C		
	Sodium	0.37 g.l ⁻¹	Liquid / 10 min	>4.5 log	
	dichloroisocyanuric acid		20°C		
	Chlorine dioxide	5%	Liquid / 10 min 20°C	3-3.3 log	
	Sodium hypochlorite	3%	Liquid / 10 min	>4.5 log	

			20°C		
	Hydrogen peroxide	Up to 50% (m/v)	Liquid / 15 min 20°C	3 log (1.56%)	Martin and Maris, 2012
Aspergillus westerdijkiae	Benzalkonium chloride	0.3-5%	Surface / 15 min / 20°C	0.1-1.9 log	Bernardi et al., 2021
	Biguanide	0.3-5%	Surface / 15 min / 20°C	0.1-0.9 log	
	Peracetic acid	0.15-3%	Surface / 15 min / 20°C	0.1-2.9 log	
	Sodium hypochlorite	0.01-1%	Surface / 15 min / 20°C	0->5 log	
	Ortho-phenylphenol	1g/m^3	Fumigation	2-3.9 log	
	Peracetic acid	0.3-1%	Surface / 15 min	1->5 log	Gonçalves Lemos <i>et al.</i> , 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-4.9 log	
	Iodine	0.2-1%	Surface / 15 min	1->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-3.9 log	
Chaetomium globosum	Peracetic acid	0.1%	Surface / 40°C	4 log (1-3h)	Scaramuzza <i>et al.</i> 2020
Cladosporium cladosporioides	Benzalkonium chloride	2-5%	Liquid / 15 min	2->4 log	Bernardi et al., 2018
	Biguanide hexamethylene	2-5%		<1 log	
	Peracetic acid	0.15-3%		1->4 log	
	Quaternary ammonium	0.3-5%		1->4 log	
	Sodium hypochlorite	0.1-1%		>4 log	
	Sodium hypochlorite	1-3 mg.L ⁻¹ free chlorine	Liquid pH 7; 25°C	2 log (139 mg.min.l ⁻¹)	Pereira et al., 2013
Cladosporium herbarum	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	4.1 log (5 min)	Reynolds et al., 2012
Cladosporium tenuissimum	Sodium hypochlorite	1-3 mg.l ⁻¹ free chlorine	Liquid pH 7; 25°C	2 log (71 mg.min.l ⁻¹)	Pereira et al., 2013
Cladosporium sp.	Chlorine dioxide	0.5-3 mg.l ⁻¹	Liquid	$1 \log (1 \text{ mg.min.l}^{-1})$	Wen et al., 2017

			pH 6, 7/10, 27°C		
	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2.9->4.1 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>4.1 log	
	Ethanol	70%	Liquid / 10 min 20°C	>4.1 log	
	Isopropanol	70%	Liquid / 10 min 20°C	>4.1 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	2.9 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>4.1 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.1 log	
Dothiorella gregaria	Chlorine dioxide	3-7 mg.l ⁻¹	Surface / 1-15 min	1.5-5 log (15 min)	Chen and Zhu, 2011
Eurotium repens (=A. pseudoglaucus)	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	3.3->4.5 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	3.3 log	
	Ethanol	70%	Liquid / 10 min 20°C	3 log	
	Isopropanol	70%	Liquid / 10 min 20°C	2.3 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	0.3 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>4.5 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.5 log	
Fusarium tricinctum	Chlorine dioxide	3-7 mg.l ⁻¹	Surface / 1-15 min	1.4-4.5 log (15 min)	Chen and Zhu, 2011

Geotrichum	Hydrogen peroxide	Up to 50%	Liquid / 15 min	3 log (0.39%)	Martin and Maris, 2012
Canalaum Ilumhonishia huntonii	Dangallyanium ahlarida	(ΠV)	20 C	0.2.10.102	Domondi at al. 2010
пурпорісній бигіони	Diguarida	0.5-3%	Liquid / 15 min	0-3.19 l0g	Bernardi <i>et al.</i> , 2019
	Demonstin a sid	2-3%	Liquid / 15 min	0.84-0.98 log	
	Peracetic acid	0.15-3%	Liquid / 15 min	0-5.67 log	
	Quaternary ammonium	0.3-5%	Liquid / 15 min	0-2.35 log	
	Sodium hypochlorite	0.01-0.2%	Liquid / 15 min	0-1.89 log	
Monascus ruber	Quaternary ammonium	1.5, 2%	Liquid / 10 min	2.9->4.1 log	Bundgaard and Nielsen 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>4.1 log	
	Ethanol	70%	Liquid / 10 min 20°C	1.3 log	
	Isopropanol	70%	Liquid / 10 min 20°C	0.9 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	1.6 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>4.1 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	2.9 log	
Mucor bainieri	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	5.2-5.3 log (5 min)	Reynolds et al., 2012
Neosartorya pseudofischeri	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	3.3->4.5 log	Bundgaard and Nielsen, 1995
· · · ·	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	3.3 log	
	Ethanol	70%	Liquid / 10 min 20°C	4 log	
	Isopropanol	70%	Liquid / 10 min 20°C	3.3 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	3.6 log	

	Chlorine dioxide	5%	Liquid / 10 min 20°C	3.3 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	4 log	
Paecilomyces fulvus	Benzalkonium chloride	0.3-2%	Surface / 15 min	2-3.9 log	Stefanello et al., 2020
	Biguanide	2-5%	Surface / 15 min	2-2.9 log	
	Iodine	0.2-1%	Surface / 15 min	2-4.9 log	
	Peracetic acid	0.3-1%	Surface / 15 min	3->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	2-3.9 log	
	Ortho-phenylphenol	15% 1g/m ³	Fumigation / 7h	>5 log	
Paecilomyces niveus	Chlorine dioxide	50-1000 ppm	Liquid / 5-60 min	>5 log (500 ppm/5 min)	Dijksterhuis et al., 2018
	Benzalkonium chloride	0.3-2%	Surface / 15 min	2->5 log	Stefanello et al., 2020
	Biguanide	2-5%	Surface / 15 min	2-3.9 log	
	Iodine	0.2-1%	Surface / 15 min	3-4.9 log	
	Peracetic acid	0.3-1%	Surface / 15 min	3->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	2-3.9 log	
	Ortho-phenylphenol	15% 1g/m ³	Fumigation / 7h	>5 log	
Paecilomyces variotii	Chlorine dioxide	50-1000 ppm	Liquid / 5-60 min	>5 log (500 ppm/5 min)	Dijksterhuis et al., 2018
	Benzalkonium chloride	0.3-2%	Surface / 15 min	1-3.9 log	Stefanello et al., 2020
	Biguanide	2-5%	Surface / 15 min	1-1.9 log	
	Iodine	0.2-1%	Surface / 15 min	1->5 log	
	Peracetic acid	0.3-1%	Surface / 15 min	1->5 log	
	Sodium hypochlorite	0.5-1%	Surface / 15 min	1-1.9 log	
	Ortho-phenylphenol	15% 1g/m ³	Fumigation / 7h	>5 log	
Penicillium	Sodium hypochlorite	500 ppm free	Surface	2.6 log (5 min)	Nierop Groot <i>et al.</i> ,
bialowiezense		chlorine	7°C		2019
	Chlorine dioxide	500 ppm free chlorine	Surface 7°C	>5 log (5 min)	
Penicillium	Sodium hypochlorite	500 ppm free	Surface	0.9 log (5 min)	Nierop Groot et al.,
buchwaldii		chlorine	7°C		2019
	Chlorine dioxide	500 ppm free	Surface	$>5 \log (5 \min)$	

		chlorine	7°C		
Penicillium brevicompactum	Sodium hypochlorite	1-10%	Surface / 1-30 min	>7.3 log (30 min)	Ebling, 2007
Penicillium caseifulvum	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2.7->4.9 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	2.7 log	
	Ethanol	70%	Liquid / 10 min 20°C	2.7 log	
	Isopropanol	70%	Liquid / 10 min 20°C	2.7 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	1.9 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	2.7 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.9 log	
Penicillium chrysogenum	Ethanol	5-10%	Liquid / 1-4 days 10-30°C 0.7-0.9 <i>a</i> _w	0.36-5.78 log	Dao <i>et al.</i> , 2008
	Ethanol	5-10%	Vapour / 1-4 days 10-30°C 0.7-0.9 <i>a</i> _w	0.13-2.47 log	
	Ethanol	0.67 kPa	Vapour/ 48-120 h	4-6.5 log	Dao and Dantigny, 2009
	Ethanol	0.7-7.5 kPa	Vapours / 24 h 25°C	3.1->5 log	Dao et al., 2010
	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	4.6-5.4 log (5 min)	Reynolds et al., 2012
	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	4->5.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium	2%	Liquid / 10 min	4 log	

	+ aldehydes		20°C		
	Ethanol	70%	Liquid / 10 min 20°C	>5.2 log	
	Isopropanol	70%	Liquid / 10 min 20°C	>5.2 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	3.7 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>5.2 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>5.2 log	
Penicillium citrinum	Sodium hypochlorite	1-3 mg.l ⁻¹ free chlorine	Liquid pH 7; 25°C	2 log (959 mg.min.l ⁻¹)	Pereira et al., 2013
Penicillium cyclopium	Hydrogen peroxide	Up to 50% (m/v)	Liquid / 15 min 20°C	3 log (1.56%)	Martin and Maris, 2012
Penicillium commune	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1-2.9 log	Bernardi et al., 2018
	Biguanide hexamethylene	2-5%		1-1.9 log	
	Peracetic acid	0.15-3%		2->4 log	
	Quaternary ammonium	0.3-5%		2->4 log	
	Sodium hypochlorite	0.1-1%		2->4 log	
	Benzalkonium chloride	0.3-5%	Surface / 15 min / 20°C	0.1-4 log	Bernardi et al., 2021
	Biguanide	0.3-5%	Surface / 15 min / 20°C	0-2 log	
	Peracetic acid	0.15-3%	Surface / 15 min / 20°C	0.1->5 log	
	Sodium hypochlorite	0.01-1%	Surface / 15 min / 20°C	0.1->5 log	
	Ortho-phenylphenol	1g/m ³	Fumigation	0-0.9 log	
	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2.7-4.3 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	2.7-4.3 log	
	Ethanol	70%	Liquid / 10 min	2.9-4 log	

			20°C		
	Isopropanol	70%	Liquid / 10 min 20°C	2.7-4 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	1.7-4 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	4.3->5.2 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	1.7-4 log	
	Ethanol	70%	Liquid / 1-5 min	2.35-5.4 log	Visconti et al., 2020
Penicillium coryphilum	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	3.7->4.8 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>4.8 log	
	Ethanol	70%	Liquid / 10 min 20°C	4.6 log	
	Isopropanol	70%	Liquid / 10 min 20°C	>4.8 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	4.4 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	>4.8 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.8 log	
Penicillium crustosum	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	4->5.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>4.2 log	
	Ethanol	70%	Liquid / 10 min 20°C	4 log	
	Isopropanol	70%	Liquid / 10 min 20°C	4 log	

	Sodium	0.37 g.l ⁻¹	Liquid / 10 min	2.3 log	
	dichloroisocyanuric acid		20°C		
	Chlorine dioxide	5%	Liquid / 10 min 20°C	4 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>5.2 log	
Penicillium digitatum	Ethanol	5-10%	Liquid / 1-4 days 10-30°C 0.7-0.9 <i>a</i> _w	0.76-3.19 log	Dao <i>et al.</i> , 2008
	Ethanol	5-10%	Vapour / 1-4 days 10-30°C 0.7-0.9 <i>a</i> _w	2.15-3.95 log	
	Ethanol	0.67 kPa	Vapour/ 48-120 h	4-5 log	Dao and Dantigny, 2009
	Ethanol	0.7-7.5 kPa	Vapours / 24 h 25°C	4.2->5 log	Dao et al., 2010
Penicillium discolor	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	3 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	3 log	
	Ethanol	70%	Liquid / 10 min 20°C	3 log	
	Isopropanol	70%	Liquid / 10 min 20°C	3 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	1.9 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	3 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.2 log	
Penicillium	Sodium hypochlorite	50 ppm free	Liquid / 0.5-5 min	0.2-1.9 log	Okull et al., 2006

expansum		chlorine	pH 6.3, 8.5		
	Chlorine dioxyde	3 ppm free	Liquid / 0.5-5 min	1.4-4 log	
		chlorine	pH 3.7		
	Sodium hypochlorite	500 ppm free	Surface	0.7 log (5 min)	Nierop Groot et al.,
		chlorine	7°C		2019
	Chlorine dioxide	500 ppm free	Surface	>5 log (5 min)	
		chlorine	7°C		
	Sodium	25-75 ppm free	Liquid / 1-6 min	3.5-6 log (2 min)	Salamão <i>et al.</i> , 2009
	dichloroisocyanurate- based sanitizer	chlorine			
Penicillium	Sodium hypochlorite	1-3 mg.l ⁻¹ free	Liquid	$2 \log (107 \text{ mg.min.l}^{-1})$	Pereira et al., 2013
griseofulvum	51	chlorine	pH 7; 25°C		,
Penicillium italicum	Ethanol	5-10%	Liquid / 1-4 days	0.44-3.65 log	Dao et al., 2008
			10-30°C		
			$0.7-0.9 a_w$		
	Ethanol	5-10%	Vapour / 1-4 days	1.66-4.38 log	
			10-30°C		
			$0.7-0.9 a_w$		
	Ethanol	0.67 kPa	Vapour/ 48-120 h	4-5 log	Dao and Dantigny, 2009
	Ethanol	0.7-7.5 kPa	Vapours / 24 h 25°C	4.1->5 log	Dao et al., 2010
Penicillium nalsiovense	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2->4.2 log	Bundgaard and Nielsen 1995
	Quaternary ammonium	2%	Liquid / 10 min	2.3->4.2.log	
	+ aldehvdes	- /*	20°C		
	Ethanol	70%	Liquid / 10 min	2.3-3 log	
			20°C		
	Isopropanol	70%	Liquid / 10 min 20°C	2.3-3 log	
	Sodium	0.37 g.l ⁻¹	Liquid / 10 min	1.7->4.2 log	
	dichloroisocyanuric acid		20°C		

	Chlorine dioxide	5%	Liquid / 10 min 20°C	3.3->4.2 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.2 log	
Penicillium paneum	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1.39-3.96 log	Bernardi et al., 2019
	Biguanide	2-5%	Liquid / 15 min	0 log	
	Peracetic acid	0.15-3%	Liquid / 15 min	0-5.75 log	
	Quaternary ammonium	0.3-5%	Liquid / 15 min	0.96-3.19 log	
	Sodium hypochlorite	0.01-0.2%	Liquid / 15 min	0-2.30 log	
Penicillium polonicum	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1-3.9 log	Bernardi et al., 2018
	Biguanide hexamethylene	2-5%		1-1.9 log	
	Peracetic acid	0.15-3%		1->4 log	
	Quaternary ammonium	0.3-5%		1->4 log	
	Sodium hypochlorite	0.1-1.5%		2->4 log	
	Benzalkonium chloride	0.3-5%	Surface / 15 min / 20°C	0.1-1.9 log	Bernardi et al., 2021
	Biguanide	0.3-5%	Surface / 15 min / 20°C	0-0.9 log	
	Peracetic acid	0.15-3%	Surface / 15 min / 20°C	0-1.9 log	
	Sodium hypochlorite	0.01-1%	Surface / 15 min / 20°C	0-3.9 log	
	Ortho-phenylphenol	1g/m^3	Fumigation	0.1-0.9 log	
Penicillium purpurogenum	Free chlorine	48.99-194.7 mg.min.l ⁻¹	Liquid pH 7; 22.5°C	4 log (258.9 mg.min.l ⁻¹)	Ma and Bibby, 2017
	Monochloramine	90.33-531.3 mg.min.l ⁻¹	Liquid pH 8, 22.5°C	4 log (200.6 mg.min.l ⁻¹)	
Penicillium roqueforti	Benzalkonium chloride	0.3-5%	Liquid / 15 min	1-3.9 log	Bernardi et al., 2018
	Biguanide hexamethylene	2-5%		1-1.9 log	
	Peracetic acid	0.15-3%		1->4 log	

Quaternary ammonium	0.3-5%		2-2.9 log	
Sodium hypochlorite	0.1-1%		2->4 log	
Benzalkonium chloride	0.3-5%	Liquid / 15 min	0.96-4.19 log	Bernardi et al., 2019
Biguanide	2-5%	Liquid / 15 min	0.91-1.04 log	
Peracetic acid	0.15-3%	Liquid / 15 min	2.90-5.66 log	
Quaternary ammonium	0.3-5%	Liquid / 15 min	1.22-3.83 log	
Sodium hypochlorite	0.01-0.2%	Liquid / 15 min	0.06-2.28 log	
Benzalkonium chloride	0.3-5%	Surface / 15 min / 20°C	0.1-1.9 log	Bernardi et al., 2021
Biguanide	0.3-5%	Surface / 15 min / 20°C	0.1-2.9 log	
Peracetic acid	0.15-3%	Surface / 15 min / 20°C	0.1->5 log	
Sodium hypochlorite	0.01-1%	Surface / 15 min / 20°C	0.1->5 log	
Ortho-phenylphenol	1g/m^3	Fumigation	0.1-0.9 log	
Isopropyl alcohol	Un-diluted	Liquid / 1-60 min	6 log (<2-8 min)	Korukluoglu <i>et al.</i> , 2006
Peracetic acid	0.1, 0.3%	Liquid / 1-60 min	$6 \log (0.3\% / \ge 55 \min)$	
Iodine	0.5, 1%	Liquid / 1-60 min	6 log (1% / 5-16 min)	
Formaldehyde	0.2;0.5%	Liquid / 1-60 min	6 log (1-25 min)	
Quaternary ammonium compounds	0.5-2%	Liquid / 1-60 min	6 log (1% / <1-21 min)	
Chlorine	0.5-2%	Liquid / 1-60 min	6 log (0.5% / <1- min)	
Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	1-4 log	Bundgaard and Nielsen, 1995
Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	1.7-4 log	
Ethanol	70%	Liquid / 10 min 20°C	3.4-3.6 log	
Isopropanol	70%	Liquid / 10 min 20°C	3.5->5.2 log	
Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	0.9-2.7 log	
Chlorine dioxide	5%	Liquid / 10 min	3.5-4 log	

			20°C		
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>5.2 log	
	Peracetic acid	0.3%	Liquid / 10 min 20°C	0.3 log	
	Hydrogen peroxide	3%	Liquid / 10 min 20°C	0.3 log	
Penicillium solitum	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	3.7->4.8 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	3.7 log	
	Ethanol	70%	Liquid / 10 min 20°C	3.2 log	
	Isopropanol	70%	Liquid / 10 min 20°C	3.7 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	3.7 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	3.7 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.8 log	
Penicillium verrucosum	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	2.1->4.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	3.1 log	
	Ethanol	70%	Liquid / 10 min 20°C	3.1 log	
	Isopropanol	70%	Liquid / 10 min 20°C	3.1 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	2.4 log	
	Chlorine dioxide	5%	Liquid / 10 min	3.1 log	

			20°C		
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.2 log	
Penicillium sp	Chlorine dioxide	$0.5-3 \text{ mg.l}^{-1}$	Liquid pH 6, 7/10, 27°C	$5 \log (1 \text{ mg.min.l}^{-1})$	Wen <i>et al.</i> , 2017
Phoma glomerata	Sodium hypochlorite	1-3 mg.l ⁻¹ free chlorine	Liquid pH 7; 25°C	2 log (152 mg.min.l ⁻¹)	Pereira et <i>al.</i> , 2013
Scopulariopsis brevicaulis	Quaternary ammonium	1.5, 2%	Liquid / 10 min 20°C	>4.2 log	Bundgaard and Nielsen, 1995
	Quaternary ammonium + aldehydes	2%	Liquid / 10 min 20°C	>4.2 log	
	Ethanol	70%	Liquid / 10 min 20°C	>4.2 log	
	Isopropanol	70%	Liquid / 10 min 20°C	>4.2 log	
	Sodium dichloroisocyanuric acid	0.37 g.l ⁻¹	Liquid / 10 min 20°C	>4.2 log	
	Chlorine dioxide	5%	Liquid / 10 min 20°C	3 log	
	Sodium hypochlorite	3%	Liquid / 10 min 20°C	>4.2 log	
Stachybotrys chartarum	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	4.1 log (5 min)	Reynolds et al., 2012
Talaromyces bacillisporus	Peracetic acid	0.1%	Surface / 40°C	4 log (400-1200 s)	Scaramuzza <i>et al</i> . 2020
Talaromyces macrosporus	Chlorine dioxide	50-1000 ppm	Liquid / 5-60 min	>5 log (500 ppm/30 min)	Dijksterhuis et al., 2018
<i>Trichoderma</i> sp	Chlorine dioxide	0.5-3 mg.l ⁻¹	Liquid pH 6, 7/10, 27°C	3 log (1 mg.min.l ⁻¹)	Wen <i>et al.</i> , 2017
Trichophyton mentagrophytes	Sodium hypochlorite	2.4%	Spray on surface / 5-10 min	6.6 log (5 min)	Reynolds et al., 2012