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Efficacy of radiant catalytic ionization to reduce bacterial populations in air and on different surfaces

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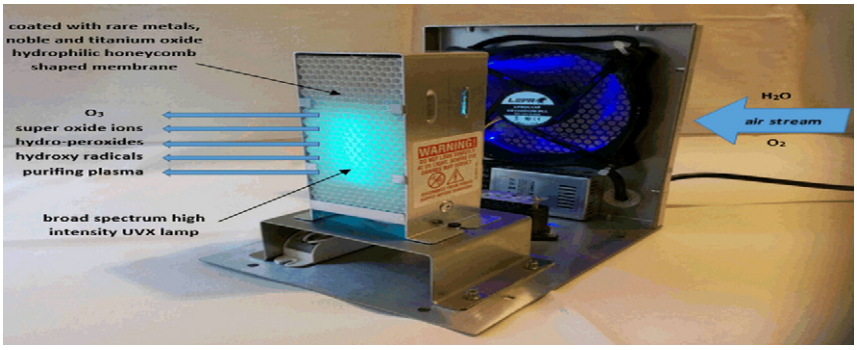
HIGHLIGHTS

- The radial catalytic ionization biocidal efficiency (RCI) has been evaluated.
- The coefficient of microbial elimination from the air was >95% after RCI usage.
- RCI usage causes visible elimination of microorganisms from tested surface.
- RCI biocidal efficiency depends on strain and type of surface.
- Spores of *Clostridium* spp. were more resistant than vegetative form of bacteria and fungi.

GRAPHICAL ABSTRACT



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ABSTRACT

Air contamination by biological agents is often observed in medical or veterinary facilities and industrial plants. Bioaerosols may sediment and pose the surface contamination. Microorganisms present on them may become a source of infections among humans and food contamination. This study determined the use of oxidative gases, including ozone and peroxide, generated by the Radiant Catalytic Ionization (RCI) cell for the inactivation of *Acinetobacter baumannii*, *Escherichia coli*, *Enterococcus faecalis*, *Pseudomonas aeruginosa*, *Salmonella* Enteritidis, *Listeria monocytogenes*, *Staphylococcus aureus*, *Streptococcus epidermidis*, *Bacillus subtilis*, *Clostridium sporogenes*, *Candida albicans*, *Aspergillus niger* and *Penicillium chrysogenumon* in air and on different surfaces. Results showed that oxidative gases produced by the RCI cell reduced all tested microorganisms. The full elimination of studied microorganisms from the air was obtained for *E. coli* and *C. albicans*. RCI also proved to be an effective method of eliminating microbes from the examined surfaces. Regarding of the species, strains origin and the type of surface, the reduction rate ranged from 19.0% for *C. albicans* to over 99% for *A. baumanii*. For both, air and surface, the most resistant to RCI was *C. sporogenes* spores, for which the percentage reduction rate ranged from –2.6% to 71.2% on the surfaces and was equal 71.7% in the air.

1. Introduction

One of the major problems of air quality is the presence of microorganisms in it, which include bacteria, molds, and viruses.

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Microorganisms present in the air compose bioaerosol. In this form microorganism may spread over considerable distances, sediment on different surfaces and causing their contamination. Ventilation ducts can be the source of airborne microbial communities (Hayleeyesus and Manaye, 2014; Hospodsky et al., 2012). Air contamination by biological agents is often observed in medical or veterinary facilities, industrial plants (e.g. food processing plants, waste segregation and recycling plants, steel and ironworks) and in agriculture (Brewczyńska et al., 2015; Douwes et al., 2003). Workers dealing with industrial waste recycling or production of highly purified biological substances are exposed to high concentrations of bioaerosols (Rim and Lim, 2014). Most particles of biological aerosols range from nanometric (e.g. bacterial endotoxins), to submicrone (e.g. fragments of bacterial or fungal cells), to particles whose diameter may exceed 100 μm (e.g. plant pollens) (Douwes et al., 2003; Dutkiewicz et al., 2011; Górny, 2010). Bioaerosols with a diameter of 1.0–5.0 μm usually remain in the air, whereas the fraction of large molecules descend on surfaces (Gąska-Jędruch and Dudzińska, 2009). It was proven that in houses the level of bacterial aerosol amounts on average to 10^3 CFU × m^{-3} (Colony Forming Units), and in workplaces 10^2 CFU × m^{-3} (Pastuszka et al., 2000).

Many surfaces, such as stainless steel, plastic, rubber org glass, are used in hospitals or food industry. Microorganisms present on them may become a source of infections in people and food contamination (Bagge-Ravn et al., 2003). The degree of surface contamination depends on their properties, such as the material of which they are made, porosity, hydrophobicity/hydrophilicity, etc. (Ismaïl et al., 2013). Schlegelová et al. (2010) indicated that surfaces that come into contact with food may be contaminated with such bacteria as *Listeria monocytogenes*, *Salmonella* spp., *Escherichia coli*, *Bacillus* spp., *Staphylococcus* spp., *Enterococcus* spp.

Ozone or UV-C radiation are commonly used for disinfection of rooms and air. (Kim et al., 1999; Kujundzic et al., 2006). It is necessary to search for and develop new methods for sterilization of air and surfaces which come into contact with patients or food. The technology of radiant catalytic ionization (RCI) is still not well known, but its popularity is gradually increasing. This technology uses the appropriate wavelength and the phenomenon of photooxidation in the presence of UV radiation and appropriate photo-catalysts, such as TiO_2, which compose the hydrophilic coating of surface of matrixes in the RCI module (Grinshpun et al., 2007). This leads to production of superoxide ions and hydroxides, and to generating plasma based on hydrogen peroxide (Cho et al., 2005). In contrast to passive methods of air purification which filter mechanically the air that flows through them, the RCI technology purifies the air outside. The advantage of this solution is the ability to perform constant disinfection of ventilated air, e.g. in food processing plants. In comparison with the effect of UV-C lamps, this technique reduces microbiological contamination and removes odors, and the resulting chemical compounds may sediment on the surface and have microbiocidal effect. It is advantage of these method, but biproducts, which are likely generated when using RCI, are of health concern. United States Environmental Protection Agency (EPA) indicate that ozone generators are not always safe and effective in removing pollutants. Harmful effects can occur following short-term exposure to low levels of ozone. Ozone generators should never be used around the ill, infirm, young or elderly people (US EPA, 1996a, 1996b). However, manufacturers of this type of air purification decelerate their safety for consumers. RCI generates very low level ozone and in the catalytic process breaks ozone down forming other oxidation products. Some authors suggest that low concentration of ozone have no effect on biological contamination (Dyas et al., 1983; Foarde et al., 1997). In this technology ozone is reduced odor, smoke and a wide spectrum of impurities in the air. Biological contamination is reduced by photocatalytic reaction with several other oxidizers. Photo catalytic oxidation must not produce any bi-products of the oxidation reaction. The EPA report confirmed the absence of bi-products using several methods, including gas chromatography, compound-specific detector tubes, and individual gas sensors (US EPA, 2000).

There are few studies in the literature assessing the efficacy of new methods for air and surface purification, such as RCI, therefore it is reasonable to conduct research on their efficacy. The study aimed to assess the efficacy of RCI towards selected microbial species present in the air and on selected solid surfaces.

2. Material and methods

The study involved the assessment of efficacy of microbial inactivation in the air and on selected types of solid surfaces as affected by radiant catalytic ionization. Ionization process was conducted using the device Induct 750 made by ActivTek Sp. z o.o, ensuring an air flow rate of 6 m × s^{-1}. The experiment was carried out in three replications for each studied strain.

2.1. Efficacy of air purification using radiant catalytic ionization

Material used for the study consisted of reference strains of bacteria (*Staphylococcus aureus* ATCC 25213, *Staphylococcus epidermidis* ATCC 35984, *Enterococcus faecalis* ATCC 29212, *Escherichia coli* ATCC 25922, *Bacillus subtilis* ATCC 8159, vegetative forms and spores of *Clostridium sporogenes* ATCC 19404) and fungi (*Candida albicans* ATCC 90028, *Aspergillus niger* ATCC 9142 and *Penicillium chrysogenum* ATCC 10106).

For the study, standardized microbial suspensions in saline with an optical density of 0.5 in McFarland standard were prepared. Next, 4 ml of each suspension was placed in a sterile nebulization chamber of the Medbryt MONSUN MP1 pneumatic inhaler. Nebulization was conducted until the complete removal the suspension from the nebulization chamber of the inhaler (about 15 min).

Nebulization chamber was placed in the testing room which was a hermetically sealed chamber with a capacity of 1.4 m^3 made of steel plates. Prior to each nebulization, the chamber walls were chemically disinfected with a preparation used for disinfection of solid surfaces, and the air contained in it was subjected to the action of the UV-C Philips TUV 36 W/G36 T8 lamp for 20 min. After that time the chamber was opened for about 20 min in order to remove the accumulated ozone. Prior to nebulization, the follow-up assessment of air microbiological purity was performed, to check the initial level of microbiological contamination. The detailed arrangement of the experiment is presented in Fig. 1, and the appearance of the research set in Fig. 2.

Air samples were collected with the compaction method using the device MAS-100 Eco (EMD Chemicals). To assess air microbiological purity after the use of UV-C Philips TUV 36 W/G36 T8 lamp and radiant catalytic ionization, 0.2 and 0.5 m^3 were collected. To assess the air microbiological contamination level in the chamber after nebulization of the microbial suspension, 0.014 and 0.052 m^3 were collected. The list of media used in the study for individual microorganisms and incubation conditions were presented in Table 1.

Colonies grown on media were counted and expressed in CFU × m^{-3} of air. Next the median was calculated for all the media studied for the given microorganism and collected air volumes. The effectiveness was expressed by giving the number of CFU of bacteria and fungi before and after the use of radiant catalytic ionization, as well as calculation of the percentage reduction rate (R[%]) according to the formula:

$$R[\%] = \frac{A-B}{A} \times 100$$

where: A – the output number of microorganisms after nebulization or drying of suspension on the solid surface [CFU × m^{-3}]B – the number of microorganisms after the use of device [CFU × m^{-3}]

Positive control in the experiment was a measurement of the number of microorganisms in the air made at 20 min after nebulization

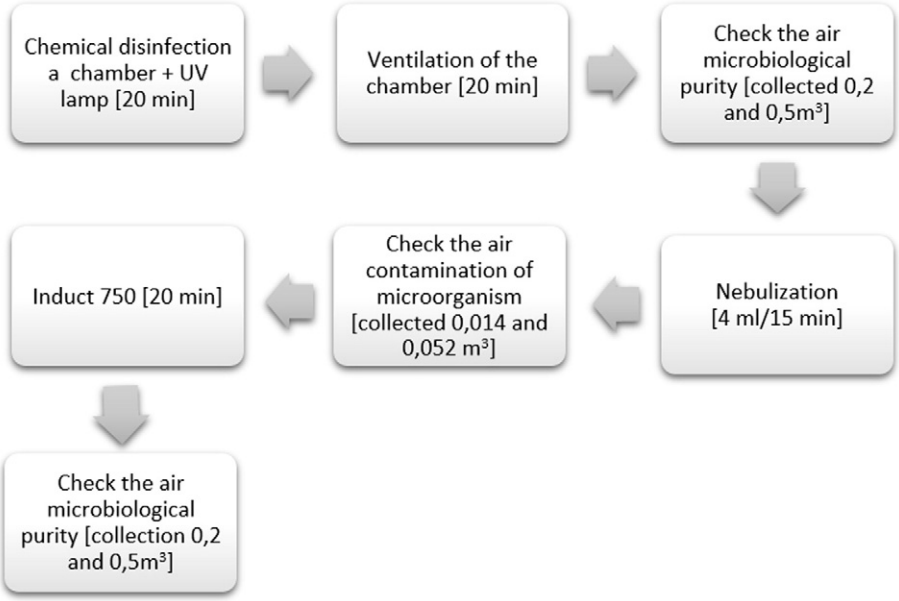

Fig. 1. Detailed arrangement of the experiment concerning the air disinfection effectiveness.

without the operation of the Induct 750. The negative control was air samples taken after preparation of the chamber prior to nebulization.

Statistical analysis of the obtained results was carried out with STATISTICA 12 PL. The calculated values of the percentage reduction rate were analyzed. Shapiro-Wilk test was carried out for the normality of distribution evaluation. The variance analysis was carried out using the one-way ANOVA and the statistical significance of the difference between the percentage reduction coefficients, based on the Tukey post-hoc parametric test at significance level $\alpha = 0.05$, were checked. As an independent variable, the microbial species was considered.

2.2. Assessment of surface disinfection effectiveness with the use of radiant catalytic ionization

Material for the study included reference strains of bacteria (*Acinetobacter baumannii* ATCC 19606, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Salmonella* Enteritidis ATCC 13076, *Listeria monocytogenes* ATCC 19111, *Staphylococcus aureus* ATCC 25213, spores of *Clostridium sporogenes* ATCC 19404) and fungi (*Candida albicans* ATCC 90028, *Aspergillus niger* ATCC 9142 and *Penicillium chrysogenum* ATCC 10106) with different biological properties. Moreover, also one clinical strain and one environmental strain were used.

In order to conduct the study, standardized microbial suspensions in 0.5% sterile solution of bovine albumin with an optical density of 0.5 in McFarland standard were prepared. Then 50 μl of each suspension were placed individually on the each studied surface and leave in the lamination chamber until complete drying.

The following solid surfaces were used in the study: steel AISI 304, polypropylene, glazed milled rock tiles, lacquered veneer, rubber and polyamide fitted carpet. Each surface had a size 1 cm × 2 cm. All surfaces before the use in the experiment were sterile.

After drying of the microbial suspension, the surfaces were inducted into the test room (photo 1) and placed at a distance of 1 m from the Induct 750 device, which was turned on for 20 min. After this time,

Fig. 2. The set for assessment of air disinfection effectiveness.

Table 1
Microbiological media used in the study and culture incubation conditions.

Microorganism	Culture media	Incubation conditions
Acinetobacter baumannii	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) MacConkey Agar (Becton Dickinson)	Aerobic 37 °C/24 h
Escherichia coli	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) MacConkey Agar (Becton Dickinson)	
Enterococcus faecalis	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) Enterococcosel Agar (Becton Dickinson)	
Pseudomonas aeruginosa	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) PYA Agar with cetrymide (Becton Dickinson)	
Salmonella Enteritidis	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) XLD Agar (Becton Dickinson)	
Listeria monocytogenes	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) ALOA Agar (Merck)	
Staphylococcus aureus *Streptococcus epidermidis*	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) Chapman Agar (Becton Dickinson)	
Bacillus subtilis	Columbia Agar with addition of 5% sheep blood (Becton Dickinson)	
Clostridium sporogenes (vegetative forms)	Columbia Agar with addition of 5% sheep blood (Becton Dickinson)	Anaerobic 37 °C/48 h
Clostridium sporogenes (spores)		
Candida albicans	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) Sabouraud Agar (Becton Dickinson)	Aerobic 25 °C/72 h
Aspergillus niger *Penicillium chrysogenum*	Columbia Agar with addition of 5% sheep blood (Becton Dickinson) Sabouraud Agar (Becton Dickinson)	Aerobic 25 °C/72 h

fragments of the surfaces were placed in a sterile solution of PBS (20 ml) and subjected to sonication in the Ultrasonic DU-4 (Nickel-Electro) sonicator, and then they were shaken for 5 min with a speed of 400 rpm (rotations per minute). The resulting suspension were 10-fold diluted using a dilution series to 10^{-4}. From each dilution, 100 μl was superficially inoculated on appropriate solid media and incubated in proper conditions (Table. 1).

Grown colonies were counted and their number was determined based on the formula:

$$L = \frac{C}{(N_1 + 0,1N_2)} * d * a$$

where: C - the total number of colonies on all plates chosen for counting,N_1 – the number of plates from the first counted dilution,N_2 – the number of plates from the second counted dilution,d – the dilution factor corresponding to the first (lowest) calculated dilution,a – the ratio of the inoculated material volume,and then was expressed in CFU per 1 cm^2 of the studied surface.

Efficacy was expressed by giving the number of CFU of bacteria and fungi before (positive control) and after the use of radiant catalytic ionization, as well as calculating the percentage reduction rate (R[%]) according to the formula given in Section 2.1.

For the obtained values of the percentage reduction rate the significance of differences were checked in the STATISTICA 12 PL software based on the post-hoc Tukey's test at the significance level $\alpha = 0,05$.

The negative control was the studied surfaces with applied sterile solution of bovine albumin.

Positive control in the experiment was the studied surfaces contaminated with tested microorganisms strains and stored without the operation of the Induct 750. The negative control was the studied surfaces with applied sterile solution of bovine albumin.

Statistical analysis of the obtained results was carried out with STATISTICA 12 PL. The calculated values of the percentage reduction rate were analyzed. Shapiro-Wilk test was carried out for the normality of distribution evaluation. The variance analysis was carried out using the multi-way ANOVA and the statistical significance of the difference between the percentage reduction coefficients, based on the Tukey post-hoc parametric test at significance level $\alpha = 0.05$, were checked. As an independent variables, the microbial strain and type of surface were considered.

Table 2
Number of microorganisms recovered from the air and percentage reduction rate R[%].

Microorganisms	Mean number of microorganisms after nebulization [CFU × m^{-3}](SE)*	Mean number of microorganisms after use of radiant catalytic ionization [CFU × m^{-3}](SE)*	Percentage reduction rate of number of microorganisms [%]
S. aureus ATCC 25213	$3,09 \times 10^5$ ($\pm 4.11 \times 10^5$)	$2,70 \times 10^1$ ($\pm 1.40 \times 10^1$)	99,99[a]
S. epidermidis ATCC 35984	$2,83 \times 10^5$ ($\pm 1.02 \times 10^5$)	$1,50 \times 10^1$ ($\pm 2.25 \times 10^1$)	99,99[a]
E. faecalis ATCC 29212	$4,11 \times 10^5$ ($\pm 2.17 \times 10^5$)	$3,50 \times 10^0$ ($\pm 5.18 \times 10^0$)	99,99[a]
E. coli ATCC 25922	$2,86 \times 10^2$ ($\pm 3.09 \times 10^2$)	n.d.	100,00[a]
B. subtilis ATCC 8159	$2,71 \times 10^5$ ($\pm 2.86 \times 10^3$)	$2,93 \times 10^3$ ($\pm 1.30 \times 10^3$)	98,92[a]
C. sporogenes ATCC 19404	$5,35 \times 10^4$ ($\pm 3.65 \times 10^4$)	$1,80 \times 10^3$ ($\pm 1.50 \times 10^3$)	96,64[a]
C. sporogenes ATCC 19404 (spores)	$4,57 \times 10^4$ ($\pm 8.00 \times 10^3$)	$1,29 \times 10^4$ ($\pm 6.60 \times 10^3$)	71,73[b]
C. albicans ATCC 90028	$4,60 \times 10^3$ ($\pm 1.81 \times 10^3$)	n.d.	100,00[a]
P. chrysogenum ATCC 10106	$9,61 \times 10^4$ ($\pm 5.37 \times 10^4$)	$3,12 \times 10^3$ ($\pm 4.72 \times 10^3$)	96,75[a]
A. niger ATCC 9142	$1,50 \times 10^5$ ($\pm 3.90 \times 10^5$)	$8,39 \times 10^2$ ($\pm 6.45 \times 10^2$)	99,44[a]

* - SE, standard error.
n.d. - not detected.
a,b – values marked with different letters are statistically significantly different ($p \leq 0.05$).

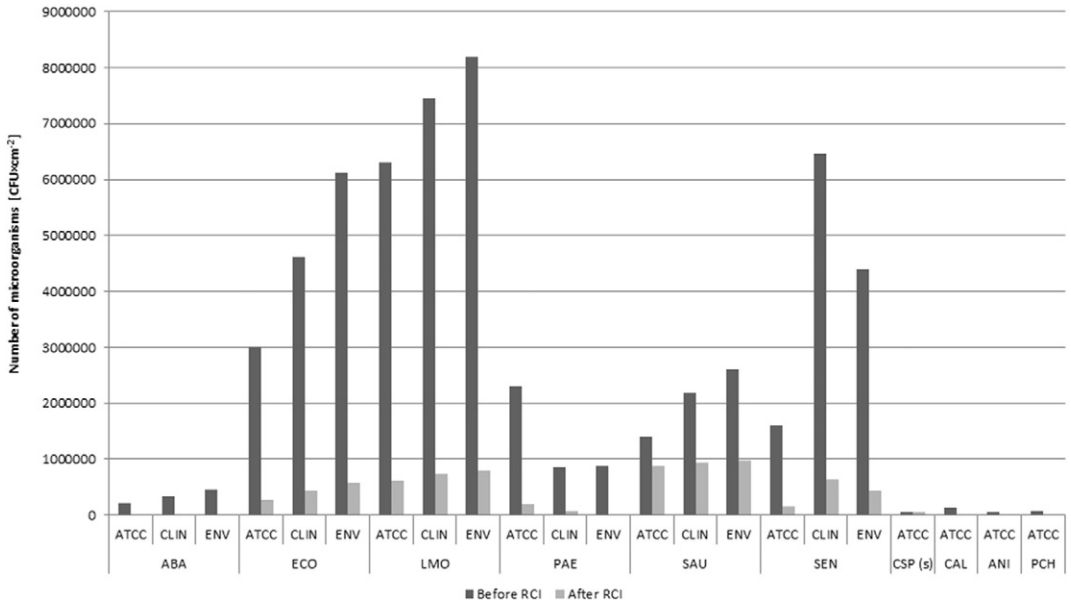

Fig. 3. Number of microorganisms recovered from steel AISI 304 before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

3. Results

3.1. Assessment of air disinfection efficacy using radiant catalytic ionization

The obtained results made it possible to determine certain fluctuation in the number of CFU of bacteria and fungi present in the air after nebulization of suspensions (Table. 2). For this reason, it was decided to introduce the absolute reduction measure in the form of R[%].

Percentage reduction rates observed for positive control ranged between 1 and 5% and therefore were not showed in the table. In samples that were negative control, the presence of microorganisms was not detected.

The using of radiant catalytic ionization caused a noticeable decrease in the number of all the studied microorganisms (Table 2).

The full elimination of studied microorganisms from the air was obtained for *E. coli* and *C. albicans*. For *S. aureus*, *S. epidermidis* and *E. faecalis* a decrease in number amounted to 4–5 logarithmic units, and the percentage reduction rate was 99.9%. Slightly lower sanitizing effectiveness of radiant catalytic ionization was noted for vegetative forms of spore-forming bacteria (R[%] – from 96.6% to 98.9%) and molds (R[%] – from 96.8% to 99.4%). The lowest effectiveness of radiant catalytic ionization was shown in relations to spores of *C. sporogenes* present in the air. The percentage reduction rate in this case amounted to 71.7% (Table 2). The demonstrated difference in the percentage reduction

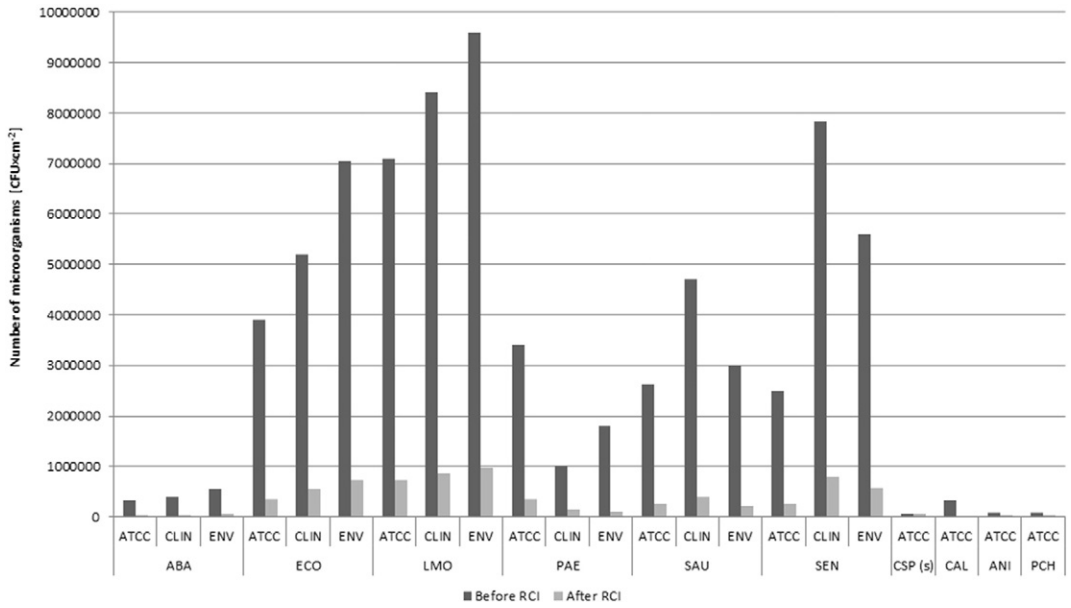

Fig. 4. Number of microorganisms recovered from polypropylene before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

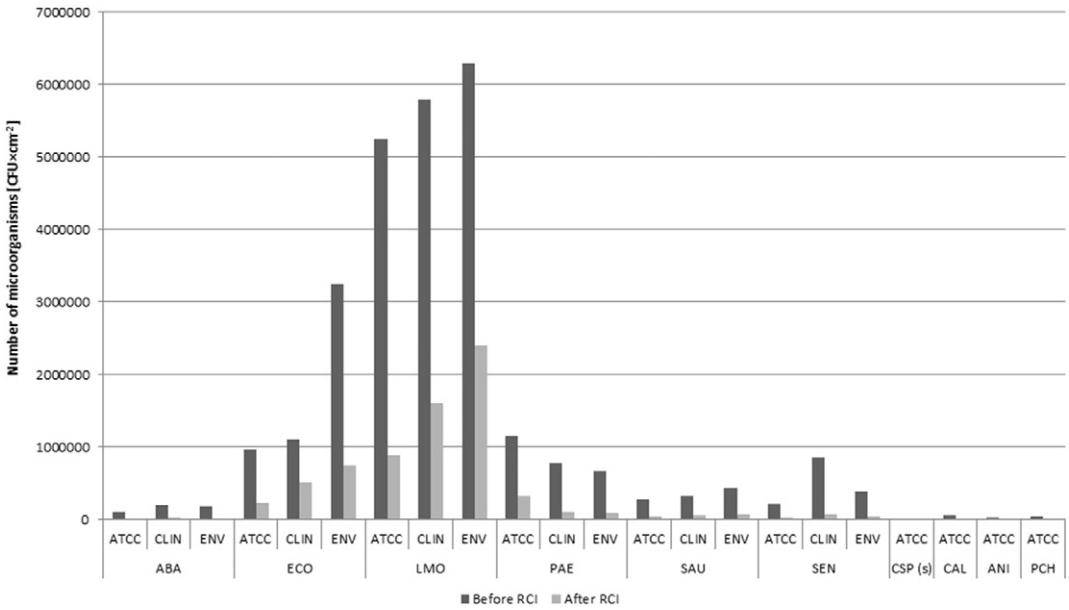

Fig. 5. Number of microorganisms recovered from glazed milled rock tiles before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

rate of the number of spores able to germinate and the number of vegetative forms of the studied microorganisms was statistically significant (Table 2).

3.2. Assessment of effectiveness of surface disinfection using radiant catalytic ionization

In positive control, any reduction of microorganisms number was not observed. In samples that were negative control, the presence of microorganisms was not detected.

The study allowed to prove a reduction in the number of bacteria as affected by radiant catalytic ionization (Figs. 3–8). Irrespective of the type of studied surface, the most resistant to the action of the used disinfection technique appeared to be *C. sporogenes* spores, for which the percentage reduction rate ranged from −2.6% on glazed milled rock tiles to 71.2% on lacquered veneer (Figs. 3–8).

The application of radiant catalytic ionization appeared to be most effective towards microorganisms present on lacquered veneer (R[%] – 71.2–99.4%) and on steel AISI 304 (R[%] – 6.6–98.9%). The lowest sanitizing effect was obtained on the polyamide fitted carpet (R[%] – 4.3–97.4%) and glazed milled rock tiles (R[%] – −2.6–90.9%) (Table 3).

Differences in the number of re-isolated bacteria resulting from different origin of the strain were observed in the case of *L. monocytogenes* applied on the surface of glazed milled rock tiles and fitted carpet. The reference strain of this species showed a statistically significantly higher reduction rate as compared with the clinical and environmental strains (R[%]: 66.93% vs. 32.86% and 31.17%, respectively), when it was applied on the surface of fitted carpet and as compared with the environmental

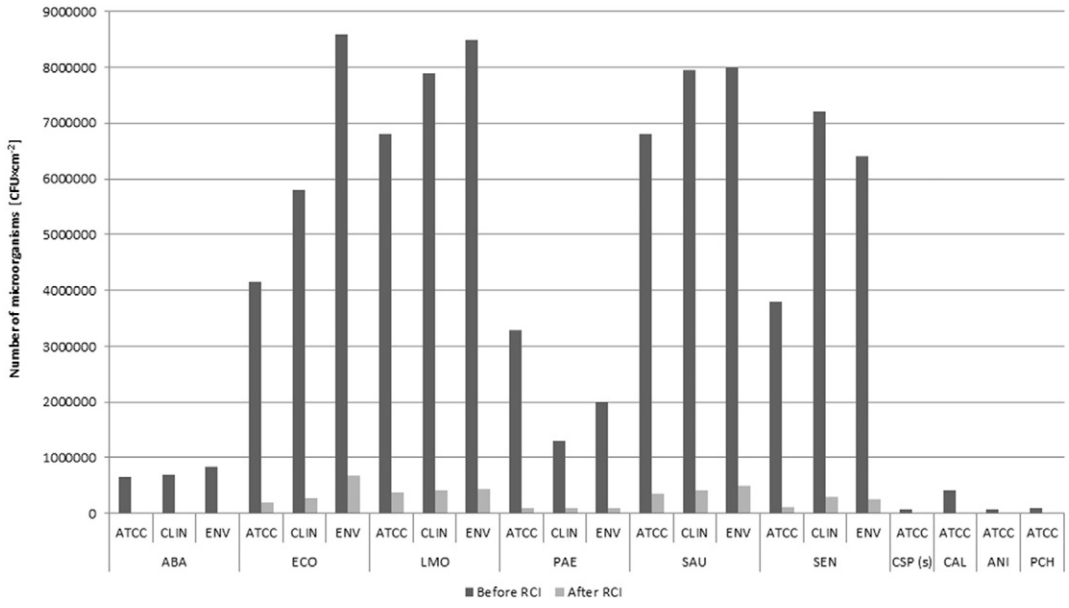

Fig. 6. Number of microorganisms recovered from lacquered veneer before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

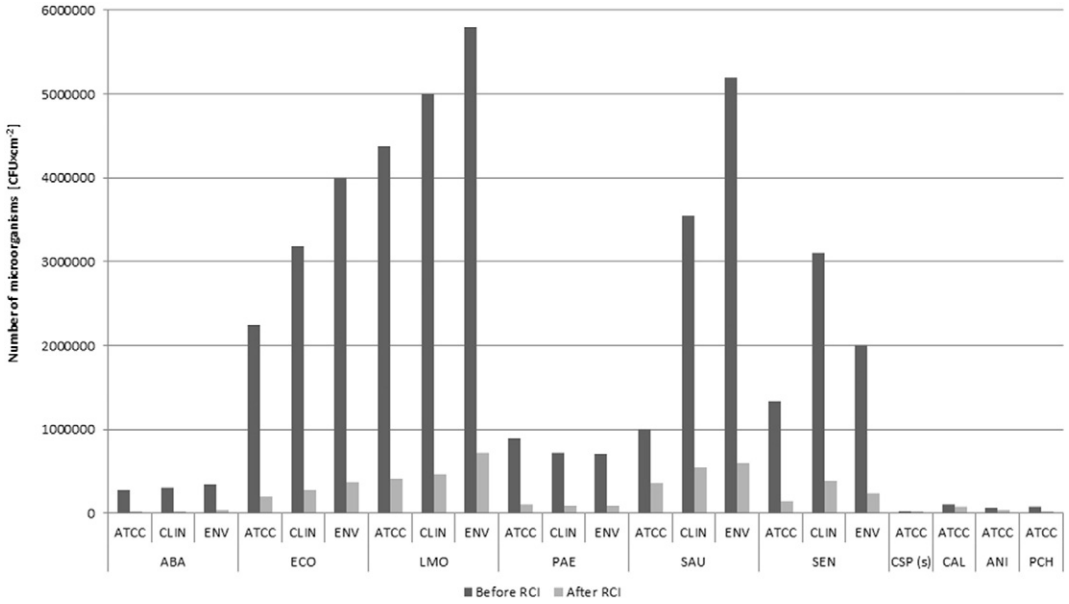

Fig. 7. Number of microorganisms recovered from rubber before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

strain (R[%] - 83.05% vs. 61.90%), when it was applied on glazed milled rock tiles. The opposite relationship was found in the case of the reference strain of *S. aureus*, which showed a statistically significantly lower reduction rate on rubber, as compared with the clinical and environmental strains (R[%] - 64.00% vs. 84.79% and 88.46%) and on steel, as compared with the environmental strain (R[%] - 37.59% vs. 62.69%) (Table 3).

Of vegetative forms of bacteria, the highest reduction rates, in the case of most studied surfaces, were obtained for *A. baumannii*. The percentage reduction rate determined for these strains stayed within the range 89.22%–99.44% (Table 3). Of the vegetative forms of bacteria, strains of *P. aeruginosa* applied on the surface of fitted carpet appeared to be the most resistant to the action of radiant catalytic ionization (R[%] - 28.00%–66.35%) (Table 3).

The studied yeast-like fungi and molds applied to surfaces of steel AISI 304 and lacquered veneer were characterized by the highest reduction rates under the influence of radiant catalytic ionization from among all the studied surfaces (R[%] 86.91%–97.3%). In most cases, the demonstrated differences were statistically significant (Table 3). Fungi *C. albicans* and *A. niger* showed the highest resistance to the action of radiant catalytic ionization on the rubber surface (R[%] = 19.00% and R[%] = 36.67%), whereas *P. chrysogenum* on the fitted carpet surface (R[%] = 42.19%) (Table 3).

4. Discussion

Disinfection of air and surfaces is necessary to reduce the risk of infections with pathogenic microorganisms in hospitals, food processing

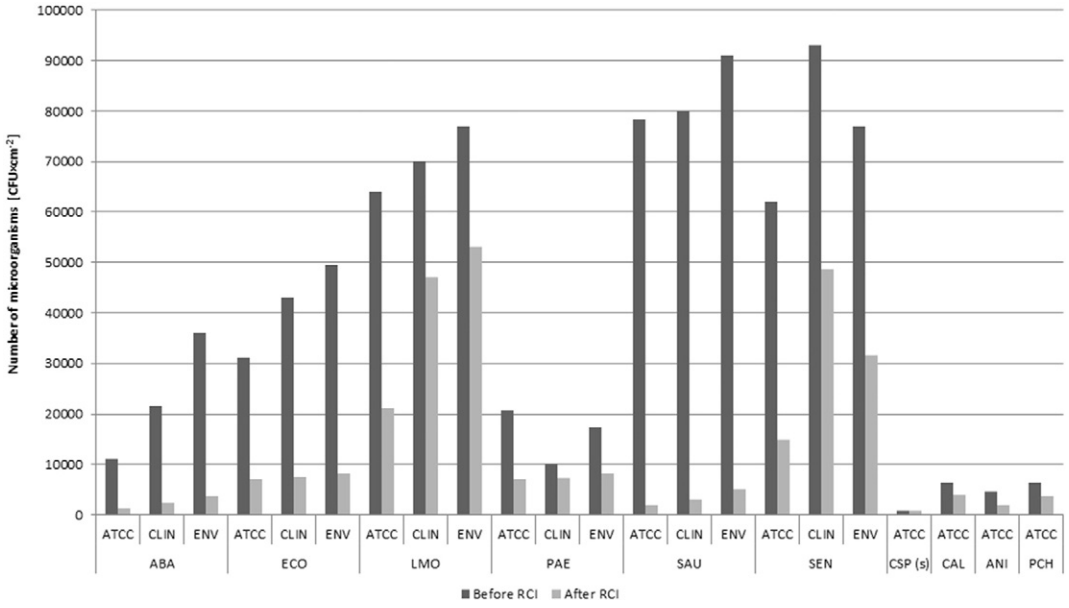

Fig. 8. Number of microorganisms recovered from fitted carpet before and after RCI (ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate).

plants, public places (schools, offices, etc.) and in houses. Ultraviolet radiation (UV) is a physical process conducted without chemicals. The source of UV-C radiation are low-pressure lamps emitting radiation with a wavelength of 253.7 nm, which inactivate bacteria, viruses, molds and spores causing DNA damage (Bintsis et al., 2000). Released atoms bind to other oxygen molecules producing ozone. It shows excellent oxidizing properties (Li et al., 2003). Artificially produced ozone is as effective as that naturally occurring, provided that its concentration is controlled. In the RCI technology ozone is not a bactericidal agent, since during the operation of the device its level does not exceed 0.05 ppm (Grinshpun et al., 2007; Ortega et al., 2007). Producer of used in this study Induct 750 declare, that this device produce ambient levels of ozone - below 0.04 ppm (ActivTek, n.d.-b, instruction of use Induct 750). This is an important advantage of the RCI technology, due to the toxicity of ozone and undesirable side effects of exposure to high ozone levels (Güzel-Seydima et al., 2004). However, EPA note that in close, non-ventilated room, level of ozone may be decrease above health concern (US EPA, 1996a, 1996b). Grinshpun et al. (2007) indicated that the generated ions (unipolar ion emission), and not photocatalytic reactions make it possible to remove contaminations such as cigarette smoke from the air. Inanimate particles are removed from the air by electrostatic precipitation, which is excited by the ionization process. Generated ions combine on the basis of electrostatic interactions with airborne contaminants, creating large and fast sediment conglomerates (Małecka and Borowski, 2011). In turn, reactive oxygen forms eliminate microbial contamination, such as bacteria, viruses or molds. Emitted chemical particles generate oxidative damage of the viral genetic material, and also impairs the functionality of the capsid protein. In bacterial cells, the coenzyme A molecules are oxidized, which results in the inhibition of cellular respiration, the oxidation of unsaturated phospholipids and the destruction of the outer membrane, and the accumulation of harmful DNA or RNA mutations (Grinshpun et al., 2007).

Sanitizing surfaces with UV-C light requires a higher dose of radiation emitting for a longer time as compared with methods utilizing the phenomenon of photo-oxidation (Ha et al., 2009; Saini et al., 2014).

The present study assessed the effectiveness of RCI towards microorganisms in the air and on different surfaces. It was proved to be the effective elimination system for many microbial species. The full elimination of microorganisms from the air was obtained for *E. coli* and *C. albicans*. The percentage reduction rate for *S. aureus*,

Table 3
Percentage reduction rate of the number of microorganisms R[%].

Strain	Percentage reduction rate [%]					
	Steel AISI 304	Polypropylene	Glazed milled rock tiles	Lacquered veneer	Rubber	Fitted carpet
ABA ATCC	98,79^{a}	90,63^{a}	89,22^{a}	99,44^{a}	90,39^{a}	89,36^{a}
	(15,12)*	(9,37)	(14.03)	(15,06)	(7,79)	(12,26)
ABA CLIN	98,82^{a}	90,75^{a}	89,35^{a}	99,30^{a}	90,53^{a}	89,40^{a}
	(15,86)	(12,27)	(10,17)	(13,17)	(10,18)	(11,48)
ABA ENV	98,86^{a}	90,34^{a}	90,86^{a}	99,39^{a}	89,91^{a}	89,44^{a}
	(13,12)	(7,39)	(11,71)	(15,22)	(12,29)	(4,16)
ECO ATCC	90,67^{a}	91,28^{a}	76,29^{a,c}	95,18^{a}	91,16^{a}	77,56^{c,e}
	(10,09)	(9,38)	(2,15)	(10,34)	(5,61)	(5,00)
ECO CLIN	90,33^{a}	89,23^{a}	53,00^{c}	95,38^{a}	91,22^{a}	82,33^{a,e}
	(12,21)	(6,45)	(1,06)	(11,61)	(8,20)	(8,03)
ECO ENV	90,52^{a}	89,65^{a}	77,16^{a,c}	92,21^{a}	90,75^{a}	83,60^{a,e}
	(11.03)	(7,18)	(5,19)	(13,07)	(11,00)	(9,14)
LMO ATCC	90,16^{a}	89,72^{a}	83,05^{a,e}	94,56^{a}	90,62^{a}	66,93^{c,e}
	(9,57)	(9,09)	(6,52)	(8,46)	(9,31)	(0,72)
LMO CLIN	90,20^{a}	89,76^{a}	72,41^{c,e}	94,75^{a}	90,80^{a}	32,86^{b}
	(12,03)	(11,56)	(2,31)	(8,26)	(7,17)	(3,11)
LMO ENV	90,37^{a}	89,84^{a}	61,90^{c}	94,94^{a}	87,67^{a}	31,17^{b}
	(9,22)	(11,63)	(1,62)	(9,28)	(13,30)	(9,67)
PAE ATCC	91,30^{a}	89,71^{a}	72,26^{c}	96,95^{a}	88,89^{a}	66,35^{c,e}
	(10,31)	(8,29)	(3,37)	(13,30)	(1,01)	(11,19)
PAE CLIN	90,75^{a}	84,00^{a}	87,01^{a}	93,08^{a}	87,64^{a}	28,00^{b}
	(10,14)	(3,15)	(3,38)	(10,14)	(7,38)	(8,07)
PAE ENV	90,23^{a}	94,44^{a}	86,02^{a}	95,00^{a}	87,07^{a}	52,02^{b,c}
	(11.19)	(9,30)	(6,21)	(10,44)	(4,17)	(7,77)
SAU ATCC	37,59^{b}	89,69^{a}	82,75^{a,e}	94,65^{a}	64,00^{c}	97,44^{a}
	(10,29)	(12,13)	(10,11)	(11,19)	(12,52)	(12,38)
SAU CLIN	56,88^{b,c}	91,70^{a}	80,94^{a,e}	94,76^{a}	84,79^{a}	96,08^{a}
	(11,07)	(10,55)	(8,18)	(12,33)	(8,22)	(8,15)
SAU ENV	62,69^{c,e}	92,67^{a}	81,61^{a,e}	93,75^{a}	88,46^{a}	94,40^{a}
	(9,47)	(8,12)	(3,46)	(10,37)	(10,52)	(6,06)
SEN ATCC	90,38^{a}	89,52^{a}	86,84^{a}	97,16^{a}	88,95^{a}	75,97^{c,e}
	(10,17)	(14,03)	(8,26)	(10,52)	(8,13)	(2,62)
SEN CLIN	90,28^{a}	89,90^{a}	90,75^{a}	95,83^{a}	87,74^{a}	47,74^{b,c}
	(10,36)	(9,21)	(6,63)	(15,02)	(13,08)	(4,84)
SEN ENV	90,23^{a}	89,59^{a}	88,30^{a}	95,95^{a}	88,00^{a}	58,83^{c}
	(10,01)	(8,28)	(8,07)	(13,20)	(10,75)	(3,06)
CSP (s)	6,61^{d}	3,23^{d}	-2,56^{d}	71,23^{c}	1,14^{d}	4,30^{d}
	(1,02)	(0,51)	(0,40)	(11,30)	(0,30)	(0,53)
CAL ATCC	92,93^{a}	97,02^{a}	88,00^{a}	96,84^{a}	19,00^{b,d}	37,50^{b}
	(13,25)	(8,03)	(7,12)	(11,59)	(13,21)	(3,17)
ANI ATCC	86,91^{a}	60,00^{c}	68,85^{c,e}	95,91^{a}	36,67^{b}	56,52^{b,c}
	(14,12)	(2,16)	(8,50)	(12,13)	(2,15)	(0,97)
PCH ATCC	90,13^{a}	53,85^{c}	75,06^{c,e}	97,30^{a}	66,71^{c}	42,19^{b,c}
	(15,52)	(1,09)	(5,88)	(11,08)	(1111)	(1,27)

ABA – *Acinetobacter baumanii*, ECO – *Escherichia coli*, LMO – *Listeria monocytogenes*, PAE – *Pseudomonas aeruginosa*, SAU – *Staphylococcus aureus*, SEN – *Salmonella* Enteritidis, CSP(S) – *Clostridium sporogenes* spores, CAL – *Candida albicans*, ANI – *Aspergillus niger*, PCH – *Penicillium chrysogenum*, ATCC – refrence strain, CLIN – clinical isolate, ENV – environmental isolate.
a,b,c, … - values marked with different letters are significantly different ($p \leq 0.05$).
* - standard deviation.

S. epidermidis and *E. faecalis* was 99.9%. Lower effectiveness of RCI was recorded in the case of vegetative forms of spore-forming bacteria (R[%] – from 96.6% to 98.9%) and molds (R[%] – from 96.8% to 99.4%). The lowest effectiveness of RCI was proved in relations to *C. sporogenes* spores present in the air (R[%] – 71.7%). Also Grinshpun et al. (2007) indicated a reduction of effectiveness of the RCI technology towards spores of *B. subtilis* in the 2.75 m^3 chamber. Approximately 75% of airborne *B. subtilis* spores exposed to the air purifier were inactivated during the first 10 min, 85% during the first 15 min, and about 90% or greater after 30 min. In present study a hermetically sealed chamber had a capacity of 1.4 m^3. The observed high rates of reduction in the number of germination able spores may be due to the fact that both experiments were carried out in relatively small cubic chambers. These results are promising for real indoor environments, where air pollution is much lower. Coverage study air purification amounts to 70 m^3 (ActivTek, n.d.-b, instruction of use Induct 750). Experiments carried in University of Agriculture in Krakow indicated that RCI was effective in rooms of cubature 20 and 45 m^3. The percentage reduction rate for total number of bacteria, *Staphylococcus* spp. and fungi ranged from 73,1% to 82,0% The results of the research carried out in University of Agriculture in Krakow about efficacy of RCI technology (Barabasz et al., 2014). This study indicated that RCI technology can be used in real rooms with different cubature. This technology was tested in museums, hospitals and hotels (ActivTek, n.d.-a, documents about use of product, http://activtek.pl/dokumenty/, accessed 10.06.2017).

In the present study it was found that the effectiveness of RCI on the surfaces was the lowest in the case of *C. sporogenes* spores, for which the percentage reduction rate ranged from −2.6% (glazed milled rock tiles) to 71.2% (lacquered veneer). One reason that spores were more resistant to RCI may be their thick membrane layer containing peptidoglycans (Berberidou et al., 2012). To the best of our knowledge the mechanism of act the RCI on spores is not fully understood. It must also be stressed that time of exposure is very important for effective inactivation of spores. In present study action time of RCI was only 20 min, which may be insufficient to kill bacteria spores. Scanning electron microscopy of *B. stearothermophilus* endospores indicated that photocatalytic oxidation affected negatively on the shape and structure of the spore. The sporicidal effect of oxidation is increasing over time (Berberidou et al., 2012).

The use of radiant catalytic ionization appeared the most effective in the case of microorganisms present on lacquered veneer (R[%] – 71.2–99.4%) and on steel AISI 304 (R[%] – 6.6–98.9%). The lowest effectiveness of RCI was shown on the polyamide fitted carpet (R[%] – 4.3–97.4%) and glazed milled rock tiles (R[%] – −2.6–90.9%).

On the surface of steel AISI 304, the most resistant to RCI were strains of *S. aureus* (R[%] - 37.6%–62.7%. The most susceptible were strains of *A. baumannii*, for which R[%] was almost 99%. Ortega et al., 2007 indicated >90% efficacy of RCI towards *S. aureus*, *Bacillus* spp., *E. coli*, *L. monocytogenes*, *C. albicans* on the surface of steel. In the present study, on different surfaces, such as rubber and fitted carpet, a considerably lower effectiveness of RCI towards *C. albicans* was shown (R[%] - 19%–37.5%). This may be related to the structure of the studied surfaces and their different properties. Moreover, contaminated surfaces were exposed to RCI products for only 20 min. In further experiments, the effect of exposure time on change in the number of microorganisms on the studied surfaces should be determined. Other authors indicated a decrease in the number of microorganisms along with growing time of exposure (Ortega et al., 2007). The efficacy of photooxidation towards *L. monocytogenes* on the surface of steel was also proved by Saini et al. (2014) (4.37 log CFU/coupon on stainless steel after 15-min treatment).

The application of UV-C light to reduce superficial microbial contamination of food is commonly known. The effectiveness of this method has been demonstrated to reduce contamination of fruits and vegetables and meat (Adhikari et al., 2015; Chun et al., 2010; Martínez-Hernández et al., 2015). Saini et al. (2014), also proved the effectiveness of photo-oxidation in food disinfection (ready-to-eat cheese and turkey). Microorganism reduction amounted to >2 log CFU/sample after 5 min of exposure. This is an important advantage of devices utilizing the phenomenon of photooxidation, UV light and ozone.

In the available literature there is no information concerning the disinfection of different surfaces with the RCI technique. In the present study such a comparison was made, since the RCI technology may be used in various environments, both in industry, where elements of steel and rubber predominate, and in everyday life, in houses, offices, etc., where there are more fitted carpets or surfaces made of lacquered veneer.

The present study proves that the RCI technology is an effective method for air and surface disinfection, although its effectiveness is varied depending on the microbial species. The possibility of widespread use of devices utilizing RCI, even when there are some people in the room, is their undoubted advantage, which may contribute to improving working conditions both in places where the air is contaminated and also where there is a risk of surface contamination.

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