BACTERICIDAL ACTION OF THE PLASMA OF HIGH-FREQUENCY CAPACITIVE AND BARRIER DISCHARGES ON MICROORGANISMS

V. V. Azharonok,^a L. E. Krat'ko,^a Ya. I. Nekrashevich,^a I. I. Filatova,^a L. A. Mel'nikova,^b N. V. Dudchik,^b S. A. Yanetskaya,^b and M. K. Bologa^c

The bactericidal action of the plasma of a high-frequency discharge excited at a frequency f = 5.28 MHz and a low pressure in air on different test-strains of microorganisms has been investigated. The high-efficiency plasma inactivation of strains of E. coli, B. subtilis, C. albicans, and S. aureus at an initial contamination $N_0 \le 10^3$ CFU/ml was detected. It was established that the most probable sterilization agents of the plasma generated are the "hot" and "cold" OH radicals, the excited electrically neutral N_2 and O_2 molecules, and the UV plasma radiation.

Keywords: high-frequency discharge, plasma, microorganisms, inactivation.

Introduction. The development of new highly efficient methods for sterilization of materials is of profound importance for medicine, ecology, and light and food industries. At present, the traditional methods of sterilization, vapor sterilization and autoclaving at a sterilizing-medium temperature of $120-180^{\circ}$ C, are most generally employed. However, in a number of cases, the use of these methods leads to damage of thermolabile materials. This circumstance has stimulated the development of alternative methods of sterilization: chemical ethylene oxide and formaldehyde gas methods and physical methods in which UV radiation, electron beams, γ -radiation, and microwave electromagnetic fields are used. However, practical implementations of these methods have revealed a number of weaknesses. For example, materials sterilized with the use of chemical agents should be then treated for a long time (as long as 24 h) because these agents can be highly inflamable, explosive, toxic, or carcinogenic substances dangerous for the environment. The sterilization of materials by γ -radiation or electron beams can cause damage to them and calls for the use of expensive, cumbersome equipment and special protective measures for provision of safety of the attending personnel. The use of high-intensity microwave electromagnetic fields is limited because a long time is required to attain a sterilization action with these fields.

Of special importance is the sterilization problem in modern medicine, in which new expensive materials and high-tech equipment are used. These circumstances as well as the discovery of new stable forms of pathogenic microorganisms impose stringent requirements on the sterilization process and call for the development of inexpensive, safe and rapid methods for treatment of materials and instruments with the use of new highly efficient sterilizing agents exerting a nondestructive action on their surface. The plasma methods of sterilization most fully satisfy these requirements [1, 2].

The suggestion that plasma can be used for sterilization of medical materials and instruments was made as early as in the 1960s of the past century [3]. This plasma can be generated by different electric discharges. Electric (arc, spark, and corona) discharges excited at atmospheric pressure are sources of plasma for sterilization of liquid objects [4–6], and the surface of solid bodies is treated by the plasma of glow discharges [7, 8]. High-frequency capacitive discharges (HFCD) of low pressure are the most suitable sources of plasma for sterilization of products made from capillary-porous materials (fabric, paper, board, atraumatic sutural material, porous fluoroplastic, etc). These discharges, unlike the other forms of gas discharges, make it possible to perform volume treatment of materials because

^aB. I. Stepanov Institute of Physics, National Academy of Sciences of Belarus, 68 Nezavisimost' Ave., Minsk, 220072, Belarus; email: azharonok@imaph.bas-net.by; ^bRepublican Scientific-Practical Center of Hygiene, 8 Academicheskaya Str., Minsk, 220013, Belarus; ^cInstitute of Applied Physics, Academy of Sciences of Moldova, Kishinev. Translated from Inzhenerno-Fizicheskii Zhurnal, Vol. 82, No. 3, pp. 425–432, May–June, 2009. Original article submitted March 26, 2008.



Fig. 1. Diagram of the experimental setup: 1) upper electrode; 2) lower electrode; 3) grounded metal support; 4) dielectric spacer; 5) vacuum chamber; 6) carrier with microorganisms; 7) quartz plates; 8) condenser; 9, 10) voltmeters; 11) work-coil; 12) high-frequency generator; 13) photographic objective; 14) digital video camera.

they give rise to nonself-sustained pulse-periodic microdischarges inside their pores [9]. Electrons in the HFCD plasma possess energy of several electron-volts and are extremely overheated relative to the heavy particles with a gas temperature of ~0.3 eV, which makes it possible to obtain a large number of excited chemically active "cold" particles, the deactivation of which can be accompanied by the appearance of a photon flux with an energy of ~6 eV. Thus, the plasma serves simultaneously as a low-temperature chemically active medium and as an UV-radiation source possessing bactericidal properties. Moreover, the sterilizing agents formed in the plasma of a discharge exist only during its combustion and disappear practically instantaneously once the discharge is switched off.

The available literature data on the sterilization of materials by the plasma of high-frequency discharges are few in numbers and were obtained for discharges excited at an industrial frequency f = 13.56 MHz [10–12]. It was established in [10] that the sterilization of materials in the plasma of a high-frequency capacitive discharge excited in air is threshold in character and its main mechanism is the destruction of microorganisms by positive ions and excited molecules of the neutral gas. The authors of [11] have shown that, in the case where a high-frequency discharge excited in oxygen is used for sterilization of materials, the degradation of microorganisms in them is due to the photoetching and ionic etching caused by OI atoms and metastable molecules of the singlet oxygen O_2^* . It was established in [12] that a high-frequency discharge excited in an atmosphere of H₂, N₂, O₂, Ar or their mixtures can be used to advantage for the inactivation of microorganisms, the removal of proteins, and the depyrogenation of endotoxins and that the sterilization effect is determined first of all by the efficiency of the erosion and etching of the surface layers of materials, protecting the microorganisms from the action of UV radiation. It was also shown that O_2/H_2 mixtures are the most suitable media for both the inactivation of bacteria and the removal of endotoxins and protein films.

At present there are no works devoted to investigating the action of the plasma generated in an electromagnetic field of lower industrial frequency on microorganisms, even though it is known that the accumulation and ionization of excited chemically active molecules in an oscillating electric field is dependent on its frequency [13, 14].

The aim of the present work is to investigate the bactericidal action of the plasma of a low-pressure high-frequency planar discharge excited at a commercial frequency f = 5.28 MHz on different test-strains of microorganisms.

Experimental and Investigation Methods. The scheme of the experimental setup used in our investigations is shown in Fig. 1. A high-frequency planar volume discharge burning uniformly in the atmosphere of molecular gases at a pressure of 0.1-10 torr was excited in a discharge chamber formed by parallel plane metal electrodes 1 and 2 mounted on the transparent quartz plates 7. The distance between the electrodes l was changed from 6 to 20 mm. The lower electrode 2 was positioned on the dielectric (glass or fluoroplastic) spacer 4 fixed to the grounded metal support 3. All the elements of the discharge chamber were inside the vacuum chamber 5 filled with a working gas. The pressure of the plasma-forming gas P was controlled by a VDG-1 vacuum gauge.

As the electric-power source 12, an industrial VChI-62-5-IG-101 generator was used. The high-frequency voltage U_1 supporting the discharge was supplied from several turns of work-coil 11 of generator 12 and was fed to the upper electrode 1. This voltage was measured by a static S509 voltmeter 10 and was varied by changing the anode



Fig. 2. Video images of the excited HFCD (a) and HFBD (b, c): 1) upper electrode; 2) lower electrode; 3) light-emitting layer of the glow-discharge plasma; 4) filament of the HFBD; 5) carrier with microorganisms in the HFCD; 6) carrier with microorganisms in the HFBD.

voltage of the generator lamp triode and the tapped-coil coupling of the work-coil with the discharge chamber. The specific power supplied (the electric power consumed by a unit volume of the plasma) W was estimated by the formula

$$W = [I_{\rm d} (U_1 - U_2)]/V$$
.

The voltage U_2 was measured by a V7-26 voltmeter 9. The current I_d was calculated by the formula $I_d = 2\pi f C U_2$, where C is the total capacitance of capacitor 8 and the lower electrode 2 relative to the ground. The capacitance of capacitor 8 was selected such that the condition $C >> C_{12}$ was fulfilled, i.e., the relation $X_{2\perp} \ll X_{d,ch}$ was provided.

As the tests-strains of microorganisms, we used vegetative forms possessing characteristic morphological, cultural and physiological-biochemical features as well as good growth properties: the ATSS-8739 gram-negative *Esherichia coli* (*E. coli*), the ATSS-6633 gram-positive *Bacillus subtilis* (*B. subtilis*), the ATSS-6538 *Staphylococcus aureus* (*S. aureus*), and the ATSS-10231 yeastlike fungi *Candida albicans* (*C. albicans*). The cultures of the microorganisms being tested were grown on agar slants for 18 h; then they were washed off by a sterile physiologic salt solution and dissolved in accordance with the Farland optical turbidity standard to a working cell concentration of 10^9-10^2 CFU/ml, whereupon control seedings were made for determining the number of living cells.

The samples prepared were put on carriers 6 inert with respect to the microorganisms and the plasma (sterile plane glass plates of size 2×2 cm or the bottom of sterile opened glass Petri dishes of diameter 9.5 cm) and placed on electrode 2 of diameter 11.5 cm in the discharge chamber. In this case, the glass plates occupied less than 3% of the area of the electrode, and the Petri dishes occupied more than 70% of its surface. The form of the discharge excited in the discharge chamber was selected depending on the indicated conditions: the treatment of the samples was performed in the plasma of a high-frequency capacitive discharge (HFBD) in the literature when Perti dishes were used [15–18]. Figure 2 presents video images of a high-frequency capacitive discharge (a) and high-frequency barrier discharges at different instants of time (b, c), obtained with the use of the photographic objective 13 and camera 14 (Fig. 1).

The high-frequency capacitive discharge was excited in air at pressure $P \approx 0.4$ torr, specific power $W \approx 0.1$ W/cm³, and interelectrode space l = 20 mm. The high-frequency barrier discharge was excited at $P \approx 0.4$ -0.5 torr, $W \approx 0.1$ W/cm³, and l = 20 mm. The time t of treatment of the test-strains by the HFCD plasma was 5, 7, and 10 min, and t was 5, 7, 10, 20, and 30 min when the HFBD plasma was used. The values of the specific power supplied to the high-frequency capacitive and barrier discharges was selected such that the heating of the samples was minimum and the possibility of their chemical sterilization was excluded. It should be noted that the HFBD excited was, at the initial stage,

TABLE 1. Survival I (%) of the Test-Cultures on the Surface of a Glass Plate after Their Treatment by the Plasma of a HFCD Versus Their Initial Concentration N_0 and the Time of Plasma Treatment. The Plasma-Forming Gas is Air, the Working Pressure $P \approx 0.4$ torr, the Electrode Spacing is 20 mm, the Specific Power Consumption $W \approx 0.1$ W/cm³

Test sulture	N_0 , CFU/ml	t, min		
Test-culture		5	7	10
E. coli	$1.0 \cdot 10^{3}$	0.2	0.2	0.2
	$8.0 \cdot 10^3$	_	6.0	0.3
	$1.4 \cdot 10^4$	_	78.0	71.0
	2			
S. aureus	8.0·10 ³	88.0		36.0
	$1.8 \cdot 10^4$	88.0	83.0	83.0
B. subtilis	$1.7 \cdot 10^2$	70.0	3.0	2.0
C. albicans	$1.0 \cdot 10^{3}$	0.2	0.0	0.0

a homogeneous glow discharge (Fig. 2b) that was then changed for the filamentary one (Fig. 2c) characterized by the appearance of a "bundle" of current-carrying plasma channels moving chaotically between the electrodes [18].

To estimate the degree of natural death of the microbe cells, we made control seedings of the prepared testcultures each on the carriers being used, and then the samples that were not subjected to the plasma action were held in an air atmosphere at room temperature for the time corresponding to the plasma-treatment regulations.

In the case where glass plates were used, the number of microorganisms found on them was determined by the method of direct seeding of the culture washed off from the experimental and control samples on the surface or in the bulk of the corresponding differential-diagnostic counting medium, including the subsequent cultivation of the seeding. When Petri dishes were used, the microorganisms in both the experimental and control dishes were flooded by a corresponding counting agarose medium (the depth-cultivation method). In this case, it was possible that one may exclude the stage of washing off the microorganisms from the carriers, leaving room for incomplete washing and secondary contamination of the biological material and, as a consequence, the appearance of an additional error in the estimation of the number of cells in the colonies of microorganisms in the process of seeding.

To obtain maximum standardized results, we cultivated the *E. coli* samples in Endo medium at a temperature of 37 $^{\circ}$ C for 72 h, the *S. aureus* samples in an egg-salt agar at a temperature of 37 $^{\circ}$ C for 24 h, the *B. subtilis* samples in a meat-peptonic agar at 30 $^{\circ}$ C for 72 h, and the *C. albicans* samples in a Sabouraud medium at 24 $^{\circ}$ C for 120 h.

The efficiency of the plasma eradication of the tests-strains and their natural lethality was estimated by the degree of survival I of the microorganisms grown after the plasma treatment, and the microorganisms grown after these strains were held for the same time t in air medium. The value of I was calculated using the relation

$$I = \frac{N_t}{N_0} \cdot 100\%$$

An analysis of samples of the control seedings of the test-cultures each has shown that the percent of microbe cells that died naturally is negligible and can be disregarded when the sterilizing plasma action is estimated. This is explained by the fact that the microorganisms being tested are mesophilic in character and are found on carriers in a physiologically neutral medium (a physiologic salt solution) or in a medium favorable for them (a nutrient medium).

Results and Discussion. Results of investigations of the action of a HFCD plasma on test-cultures *E. coli*, *S. aureus*, *B. subtilis*, and *C. albicans*, carried out with the use of glass plates as carriers, are presented in Table 1. It is seen from this table that the efficiency of the bactericidal action of the plasma generated in a high-frequency capacitive discharge excited in air depends substantially on the initial concentration N_0 of the test-culture and the duration of the plasma action t on it. For example, the survival of *E. coli* was 0.2% at $N_0 \approx 1.10^3$ CFU/ml and t = 7-10min and not lower than 71% at $N_0 \approx 1.4 \cdot 10^4$ CFU/ml at t = 7-10 min. A similar dependence was obtained for *S. aureus*, even though, in this case, the bactericidal action of the plasma was weaker: after a 10-min treatment, the survival of the microorganisms was 83% at $N_0 = 1.8 \cdot 10^4$ CFU/ml and 36% at $N_0 = 8.0 \cdot 10^3$ CFU/ml. This effect is evidently due to the formation of clumps of microorganisms stuck together when they are present in large concentrations in the culture, which prevents the penetration of the sterilizing plasma agents into them.

TABLE 2. Survival I (%) of the Test-Cultures at the Bottom of a Petri Dish after Their Treatment by the Plasma of a HFBD Versus Their Initial Concentration N_0 and the Time of Plasma Treatment t. The Experimental Conditions are Identical to Those Presented in Table 1

Test-culture	N ₀ , CFU∕ml	t, min				
		5	7	20	30	
E. coli	$1.0 \cdot 10^2$	0	0	0		
	$1.0 \cdot 10^{3}$	0	0	0		
S. aureus	$1.7 \cdot 10^{3}$	—	90	88	82	
B. subtilis	$1.0 \cdot 10^{9}$		100	100	98	

The efficiency of bactericidal action of the plasma on *B. subtilis* with $N_0 = 1.7 \cdot 10^2$ CFU/ml was low when the time of treatment was equal to t = 5 min; in this case, approximately 70% of viable cells were detected. However, when the time of plasma action was increased to 7–10 min, the survival of the *B. subtilis* cells was of the order of 2–3%. For the *C. albicans* culture with $N_0 = 1.0 \cdot 10^3$ CFU/ml, a practically complete bactericidal effect was detected at all the times of action of the HFCD plasma on it.

Data on the survival of the test-cultures *B. subtilis*, *E. coli*, and *S. aureus* found at the bottom of the Petri dish subjected to the action of the plasma of a high-frequency barrier discharge for different times are presented in Table 2. These data show that the efficiency of bactericidal action of the HFBD plasma, as well as of the HFCD plasma, depends on the initial contamination of the cellular suspension. However, unlike the HFCD plasma, a complete bactericidal effect for the *E. coli* microorganisms with $N_0 = 1.0 \cdot 10^2 - 1.0 \cdot 10^3$ CFU/ml was attained even after a 5-min treatment of them by the HFBD plasma. The survival of *S. aureus* with $N_0 = 1.7 \cdot 10^3$ CFU/ml and *B. subtilis* with $N_0 = 1.0 \cdot 10^9$ CFU/ml was 80-100% even in the case where the duration of the treatment was t = 30 min. These results, along with the analogous data obtained for the HFCD plasma, (Table 1), show that *Staphylococcus aureus* is very stable to the plasma action.

To determine the conditions for efficient eradication of *Staphylococcus aureus* in the plasma of a high-frequency barrier discharge, we excited this discharge at higher pressures of the plasma-forming gas with the use of a higher specific electric power. The results obtained have shown that an increase in the plasma-forming gas pressure from 0.4 to 1.5 torr does not cause a marked decrease in the number of the *S. aureus* microorganisms — the survival of this test-culture was ~90% when it was subjected to the plasma action for 30 min. The authors of [7] also arrived at the conclusion that the efficiency of the plasma eradication is independent of the plasma pressure. The efficiency of action of the plasma of a high-frequency barrier discharge excited with the use of a higher specific power (W =0.85 W/cm³, $P \approx 0.7$ torr) on the cultures being studied was investigated in a discharge chamber with electrodes separated by 10 mm distance: the area of these electrodes was practically equal to the area of the inserted glass plates of size 2 × 2 cm. As the test-objects, we used, in addition to *S. aureus*, *E. coli* and *B. subtilis bacteria* selected for comparison. The duration of the treatment of these cultures was 5, 10, and 20 min. The data on their survival are presented in Fig. 3.

As is seen from Fig. 3, the survival of *E. coli* comprised 13.5% of the initial microbe concentration after treatment by the HFBD plasma for 5 min. The bactericidal effect was practically absent for the *S. aureus* culture subjected to the same plasma action — the survival of its cells was of the order of 94%. The efficiency of the HFBD plasma action on the *B. subtilis* culture at t = 5 min was also low: after the plasma treatment, the viable *B. subtilis* cells comprised ~40% of their initial concentration $N_0 \approx 4.4 \cdot 10^3$ CFU/ml. The results of investigations of the survival of the cultures being studied after 10 min treatment by the HFBD plasma have shown that this plasma action gives a 100% bactericidal effect for the test-strains of *E. coli* and *B. subtilis*, and the survival of the *S. aureus* test-strain was about 4% in this case. Complete eradication of all the test-cultures was obtained after 20 min plasma action.

An analysis of the data presented in Tables 1 and 2 and in Fig. 3 shows that the microbe population subjected to the plasma treatment can be conditionally divided into three characteristic groups: microorganisms that are least tolerant to the plasma action (they die within the first minutes of treatment), microorganisms that have a "normal" tolerance (they form the main mass of the population), and microorganisms that are highly tolerant to the plasma action.



Fig. 3. Survival *I* of microorganisms versus the time of their treatment in the plasma of a HFBD at a pressure $P \approx 0.7$ torr and a specific energy consumption $W = 0.85 \text{ W/cm}^3$: 1) *E. coli*; 2) *S. aureus*; 3) *B. subtilis. I*, %; *t*, min.

The available literature data on the study of the structural-functional changes in microorganisms under the action of a plasma point to the fact that death of these microorganisms happens when their cell wall breaks down, the cytoplasmic membrane unravels, and its main function — as a permeability barrier — is destroyed [19–21]. However, at present there is no clear consensus on the mechanism of the plasma destructive action on microorganisms, which is explained by the synergetics of the acting factors as well as by the fact that the mechanism of the eradication process is unclear. In accordance with the modern concepts, the main bactericidal agents of a plasma is its UV radiation of wavelength falling within the ranges $\Delta\lambda_1 \sim 160$ –220 nm and $\Delta\lambda_2 \sim 250$ –270 nm as well as the fluxes of excited atoms and molecules of oxygen, nitrogen, OH radical, ozone, and nitrogen oxides NO_x. In this case, UV radiation exerts a determining bactericidal action because its frequencies $\Delta\lambda_1$ and $\Delta\lambda_2$ correspond to the maximum of the radiation-inactivation cross section of the deoxyribonucleic acid molecules of the microorganisms and it plays a significant role in the formation of chemically active particles in the plasma [22, 23]. According to the data obtained in [7], the unexcited and charged particles have no influence on the plasma eradication.

Our earlier investigations [14] on the influence of the regimes of excitation of a high-frequency capacitive discharge with f = 5.28 MHz on the parameters of the plasma generated in this discharge have shown that the bands of the plasma radiation in the UV, visible, and near infrared regions of the spectrum ($\Delta\lambda = 300-1000$ nm) correspond to the bands of the first and second positive systems of the N₂ molecules, the bands of the first negative system of the molecular ion N⁺₂, and the OH bands. In this case, "cold" OH molecules having a rotational temperature T_{rot}^c close to the gas-kinetic temperature $T_g \sim 300$ K in the excited electron state OH($A^2\Sigma$) and "hot" molecules with $T_{rot}^h \sim 10,000$ K are present in the discharge at the same time. An analysis of the emission spectra of a high-frequency capacitive discharge in the range 225–300 nm has shown that the intensity of the plasma radiation increases slightly when its wavelength decreases, beginning at 300 nm. In our opinion this effect is due to the presence of the "tails" of molecular bands of O₂ (the Shumann–Runge system) and molecular bands of N₂ (the Lyman–Birge–Hopfield system) in this spectral range. The foregoing allows the suggestion that the bactericidal agents of the high-frequency capacitive and barrier discharges used in the present work are the excited electrically neutral N₂ and O₂ molecules, the OH radicals, and the UV radiation of the discharge plasma.

Conclusions. Our investigations have shown that the plasma of the high-frequency capacitive and barrier discharges excited at a commercial frequency f = 5.28 MHz in air at a pressure $P \sim 0.4$ torr with the use of a specific power $W \approx 0.1-0.85$ W/cm³ is a highly efficient bactericidal medium for strains of *E. coli*, *B. subtilis*, *C. albicans*, and *S. aureus* when their initial concentration $N_0 \le 10^3$ CFU/ml. At larger values of N_0 the eradication efficiency decreases because of the formation of clumps of microorganisms that stick together, which decreases the number of plasma bactericidal agents penetrating into them. It has been established that the HFBD plasma exerts a more pronounced bactericidal action on microorganisms as compared to that of the HFCD plasma, and that the eradication of all the test-cultures being investigated is independent of the pressure (0.4-1.5 torr) of this plasma.

It was shown that the most probable bactericidal agents of plasma generated in a high-frequency capacitive and barrier discharge are "hot" and "cold" OH molecules, excited N_2 and O_2 molecules, and the UV radiation of the plasma.

To analyze the mechanisms of eradication of microorganisms under the action of the HFCD and HFBD plasma in detail and formulate engineering principles of construction of a high-efficiency sterilizing plasma apparatus safe for the environmentit it is necessary to carry out additional experimental and theoretical investigations.

The authors are grateful to S. V. Goncharik for help in the preparation of materials for this article.

NOTATION

 C_{12} , capacitance of the space between the upper and lower electrodes, μ F; f, frequency, MHz; I, degree of survival of microorganisms, %; I_d , total current of the discharge, A; l, interelectrode space, mm; N_0 , initial concentration of cells of a test-strain, CFU/ml; N_t , final concentration of cells of a test-strain, CFU/ml; P, pressure, torr; t, time of treatment of test-strains by a plasma (duration of plasma action), min; T_g , gas-kinetic temperature, K; T_{rot}^h , rotational temperature of the "hot" molecules, K; T_{rot}^c , rotational temperature of the "cold" molecules, K; U_1 , high-frequency voltage, V; U_2 , voltage across the space between electrode 2 and the ground, V; V, volume occupied by the plasma, cm³; W, specific energy supplied, W/cm³; X_{21} , reactance of the circuit section, Ω ; $X_{d,ch}$, reactance of the discharge chamber, Ω ; $\Delta\lambda_1$, $\Delta\lambda_2$, $\Delta\lambda$, wavelength ranges, nm; VUV/UV, vacuum ultraviolet/ultraviolet. Subscripts: d, discharge; 0, initial value of a quantity; g, gas; h, hot; rot, rotational; c, cold; d.ch, discharge chamber.

REFERENCES

- 1. F. Poncin-Epaillard and G. Legeay, Surface engineering of biomaterials with plasma techniques, *J. Biomater. Sci. Polym. Ed.*, **14**, 1005–1028 (2003).
- P. K. Chu, J. Y. Chen, L. P. Wang, and N. Huang, Plasma-surface modification of biomaterials, *Mater. Sci. Eng.: R: Reports*, 36, Iss. 5-6, 143–206 (2002).
- 3. W. P. Menashi, Treatment of Surfaces, US Patent No. 03383163, 14 May, 1968.
- 4. K. V. Vilkov, A. L. Grigor'ev, Yu. A. Nagel', and I. V. Uvarova, Decontamination action of a high-power pulse electric discharge in water. II. Experimental results, *Pis'ma Zh. Tekh. Fiz.*, **30**, No. 7, 48–53 (2004).
- 5. Yu. S. Akishev, M. E. Grushin, V. B. Karal'nik, A. E. Monich, M. V. Pan'kin, N. I. Trushkin, V. P. Kholodenko, V. A. Chugunov, N. A. Zhirkova, I. A. Irkhina, and E. N. Kobzev, Creation of a nonequilibrium plasma in heterophase gas-liquid media at atmospheric pressure and demonstration of its potentialities for sterilization, *Fiz. Plazmy*, **32**, No. 12, 1142–1152 (2006).
- 6. E. G. Zhuk, Action of pulse electric discharges on a microbe cell, *Élektron. Obrab. Mater.*, **37**, No. 1, 57–59 (1971).
- I. A. Soloshenko, V. V. Tsiolko, V. A. Khomich, A. I. Shchedrin, A. V. Ryabtsev, V. Yu. Bazhenov, and I. L. Mikhno, Use of a low-pressure glow discharge for sterilization of medical instruments ware, *Fiz. Plazmy*, 26, No. 9, 845–853 (2000).
- 8. Xutao Deng, Jianjun Shi, and Michael G. Kong, Physical mechanisms of inactivation of *Bacillus subtilis* spores using cold atmospheric plasmas, *IEEE Trans. Plasma Sci.*, **34**, No. 4, 1310–1316 (2006).
- 9. I. Sh. Abdullin, L. N. Abutalipova, V. S. Zheltukhin, and I. V. Krasina, in: *High-Frequency Plasma Processing* of Capillary-Porous Materials in a Dynamic Vacuum [in Russian], Izd. Kazansk. Univ., Kazan' (2004), pp. 136–143.
- 10. V. A. Lisovskiy, S. D. Yakovin, V. D. Yegorenkov, and A. G. Terent'eva, Plasma sterilization in low-pressure rf discharge, *Vopr. Atom. Nauki Tekh. Plazmen. Élektr. Nov. Metody Uskor.*, No. 1, 77–81 (2000).
- 11. A. A. Bol'shakov, B. A. Cruden, R. Mogul, M. V. V. S. Rao, S. P. Sharma, B. N. Khare, and M. Meyyappan, Radio-frequency oxygen plasma as a sterilization source, *AIAA J.*, **42**, No. 4, 823–832 (2004).
- 12. F. Rossi, O. Kylian, and M. Hasiwa, Decontamination of surfaces by low pressure plasma discharges, *Plasma Process. Polym.*, **3**, Nos. 6-7, 431–442 (2006).
- 13. O. A. Ivanov and S. F. Lirin, Excitation of electronic levels of nitrogen in a low-pressure gas discharge in ultrastrong microwave fields, *Fiz. Plazmy*, **18**, Issue 1, 124–127 (1992).
- V. V. Azharonok, I. I. Filatova, V. D. Shimanovich, and L. N. Orlov, Influence of the regimes of a high-frequency capacitive discharge on the plasma parameters in gas mixtures N₂/CO₂/He, *Zh. Prikl. Spektrosk.*, 69, No. 5, 658–664 (2002).
- 15. Yu. P. Raizer, M. N. Shneider, and N. A. Yatsenko, *High-Frequency Capacitive Discharge: Physics, Experi*mental Technique, Application [in Russian], Izd. MFTI, Moscow (1995).

- 16. J. J. Shi, D. W. Liu, and M. G. Kong, Radio-frequency dielectric-barrier glow discharges in atmospheric argon, in: *Proc. 28th ICPIG*, 15–20 July, 2007, Prague, Czech Republic (2007), pp. 2211–2214.
- 17. P. Slavicek, A. Brables, V. Kapicka, M. Klima, and M. Sira, Longitudinal emission diagnostics of plasma channel in rf barrier torch discharge, *Acta Physica Slovaca*, **55**, No. 6, 573–576 (2005).
- 18. D. S. Nikandrov and L. D. Tsendlin, Low-frequency barrier discharge, *Zh. Tekh. Fiz.*, **75**, No. 10, 29–38 (2005).
- 19. M. Moisan, J. Barbeau, M.-C. J. Crevier Pelletier, N. Philip, and B. Saoudi, Plasma sterilization. Methods and mechanisms, *Pure Appl. Chem.*, **74**, No. 3, 349–358 (2002).
- S. Moreau, M. Moisan, M. Tabrizian, J. Barbeau, J. Pelletier, A. Ricard, and L'H. Yahia, Using the flowing afterglow of a plasma to inactivate *Bacillus subtilis* spores: Influence of the operating conditions, *J. Appl. Phys.*, 88, No. 2, 1166–1174 (2000).
- N. S. Panikov, S. Paduraru, R. Crowe, P. J. Ricatto, C. Christodoulatos, and K. Becker, Destruction of *Bacillus Subtilis* cells using an atmospheric-pressure capillary plasma electrode discharge, *IEEE Trans. Plasma Sci.*, 30, No. 4, 1424–1428 (2002).
- 22. M. I. Lomaev, E. A. Sosnin, V. F. Tarasenko, D. V. Shitz, V. S. Skakun, M. V. Erofeev, and A. A. Lisenko, Excilamps of barrier and capacitance discharges and their application (Review), *Prib. Tekh. Eksp.*, **49**, No. 5, 595–616 (2006).
- 23. http://rvs.itsoft.ru:8000/article/sart.html?id=314&conf_id=4.