

Efficacy of Respiratory Antimicrobial Protection Devices

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Individual protection of the respiratory tract is a key factor in preventing/stopping a potential epidemic/pandemic caused naturally or during biological agents application in war or terrorist activities. Improving the existing or introducing new appropriate personal protective equipment to the military environment would be invaluable. In order to acquire a confirmation of reliability and physiological suitability for use in the protection of military personnel in case of a declared epidemic/pandemic, the protection effectiveness and physiological suitability of three models of epidemiological masks were tested in accordance with BS EN 149: 2007 under the conditions of simulated biological air contamination. The test results and their statistical processing have shown that an epidemiological mask possesses high efficiency and physiological suitability as a protective device in the case of air contamination with aerosols of biological agents. It is recommended for use in peacetime together with the existing standard military masks

Key words: protective mask, personal protection, epidemiological protection, respiratory protection, antimicrobial protection, epidemic, bioaerosol, silver nanoparticles, test results.

Introduction

AIR contamination with biological agents is one of the major problems in human respiratory system protection. Increased content of airborne particles that contain microorganisms may be due to natural outbreaks [1-3] or use of biological weapons [4, 5]. In terms of medical emergencies, it can be assumed that the most likely usage of biological agents could be in the form of aerosols, since microorganisms spread most efficiently through the air, and the respiratory route is the most effective way of exposure [6, 7].

A choice of biological agents for a malicious/ terrorist attack is always based on an estimation of the efficiency and the parameters such as contagiousness and pandemic potential. Thus, each milliliter of nasal fluid secretions spread millions of influenza virus particles, since a 0.1 µl aerosol particle contains more than 100 virus particles, with an average diameter of 120 nm. It is supposed that a single human infectious dose (HID) of the influenza virus might be between 100 and 1000 particles [8]. In 4 out of 12 patients suffering from influenza, the presence of the virus in the exhaled air has been confirmed, and more than 87% of the exhaled particles are smaller than 1 µm in diameter [9]. This is significant, considering the foresight of the

World Health Organization (WHO) that the mortality rate of a possible pandemic caused by the avian influenza virus H5N1 could be in the range of 10% to 35% of the world population [10, 11].

According to the WHO recommendations in case of epidemics and pandemics, along with other preventive measures to protect human health, one of the key measures is the implementation of respiratory protection [12, 13]. At the individual level, respiratory protection is achieved by wearing different types of masks, half masks, respirators or other physical barriers [14]. The mask should be worn at all occasions where there is a possibility of air contamination by viruses and bacteria due to a large number of sick or even healthy individuals (healthy carriers) in enclosed spaces, public places and other places of assembly [15].

Over the last decade, disposable protective filtering half masks were used for the protection of civilians against viruses. Their main purpose is the protection of the "inside out", but during an epidemic they are used for protection in the opposite direction as well. The application of epidemiological masks is the most common, because they have a mechanism of protection based on the antiviral and antibacterial properties of filtering material. They were widely used in Asia, particularly during the SARS (Severe Acute Respiratory Syndrome) epidemic, and proved to be

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crucial for a continuous protection against this deadly disease [16, 17]. A similar epidemiological effect in protection can be expected using epidemiological masks with impregnated silver nanoparticles against the virus A/H5N1 and other circulating viruses such as A/H7N7 and A/H7N3 (in birds), and A/H1N1 and A/H3N2 (in humans), because they are similar to the SARS coronavirus [18]. With the development of nanotechnology, the filtration properties of these devices are built to a very high level [19, 20], so they have a possibility to filtrate micron and sub-micron particles [21].

The Serbian army uses a standard military mask as basic equipment for personal respiratory protection. In case of an epidemic or pandemic, either naturally occurring or caused by biological weapons, it is necessary to maintain a high operational capability of the military personnel, both in peace and wartime. The assessments of the military protective mask M3 proved that it is physiologically suitable for use in real conditions (resistance during inhalation 115.5 Pa) and that it is an effective device of protection in case of a user's exposure to biological agents contamination through respiratory organs (filtration efficiency 99.9994 %) [22].

Although in case of an epidemic or pandemic it is advised to use a protective half mask for the protection of civilian population [23], the question is whether this kind of protection is effective and physiologically more appropriate for military personnel compared to a formation device - military protective mask, especially in peacetime conditions doing daily tasks.

The aim of this study is to explore the protection effectiveness of three models of epidemiological masks for respiratory protection in terms of possible biological air contamination and to obtain a confirmation of their reliability and suitability for use in the protection of military personnel in case of a declared epidemic/pandemic by strains similar to A/H5N1 or other influenza viruses as a future interhuman respiratory infection.

Methods

Samples of protective half mask which were tested were manufactured in the company "9. Septembar", Gornji Milanovac, Republic of Serbia. The epidemiological mask which is the subject of our study is a protective half mask made of multiple layers of different filtration materials (depending on a model). It is a disposable device for personal respiratory protection with appropriate hydrophobic characteristics. Fig.1 presents the correct way to use a protective half mask.

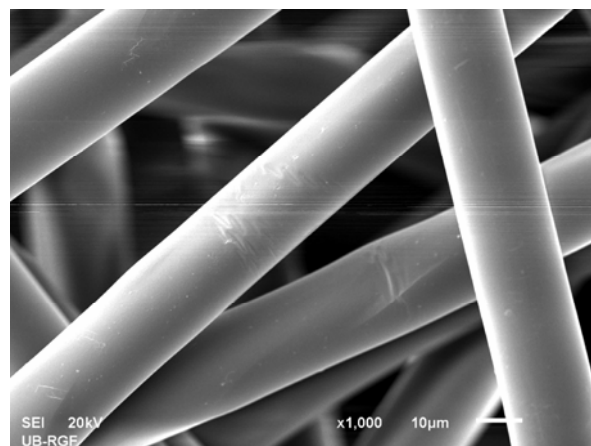


Figure 1. Epidemiological mask in a protective position

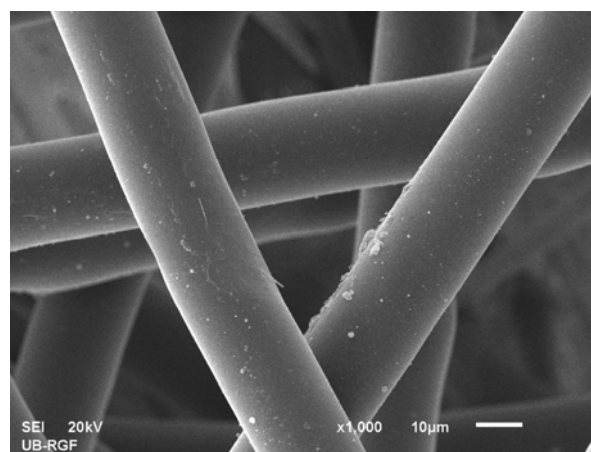
In order to improve the existing commercially available four-layered epidemiological mask, antimicrobial type, without an exhalation valve (for this testing marked as EMA-1), the same company has developed its two prototype models: a four-layered epidemiological mask, antimicrobial type, with an exhalation valve (for this testing marked as EMA-2) and a five-layered epidemiological mask, "nanotechnology" type, without an exhalation valve (for this testing marked as EMN-3). The samples of all these models were comparatively tested.

The antimicrobial nano-silver filter, which is an integral part of the model EMN-3, was produced with the basic filtration material in which 10nm silver particles with a highly developed surface were dispersed (manufacturer company "Linkason Enterprises Limited", Hong Kong). The diameter of pores for air passage into the nano-silver filter is about 3 nm.

Fig.2 shows the Scanning Electron Microscopy (SEM) images of the samples of the primary filtration material (incorporated in models 1 and 2) and the filtration material with dispersed silver nanoparticles (incorporated in model 3). The characterization of materials was done using a JSM-6610LV scanning electron microscope (manufacturer company JEOL, Japan).



a)



b)

Figure 2. SEM image of: a) primary filter material, and b) filtration material with dispersed silver nanoparticles

The most important parameters for the assessment of protection provided by the protective half mask are the determination of aerosol contaminant filtration efficiency (through the filtration material) and the total inward leakage maximum towards the interior of the half mask, with a condition of adequate inhalation resistance for physiological suitability.

Generated aerosols of a non-toxic agent were used to simulate the presence of contamination in the air [24]. Aerosols of biological agents are polydispersed systems with particles of different sizes and shapes [25]. The virus size can range from 0.015 to 0.3 μm , while the bacterial cells may range up to 2 μm [26]. Due to its physical properties, sodium chloride (NaCl) aerosol was used as a suitable simulator of biological agents. The characteristics of the particle size distribution of the generated test aerosols of NaCl are given in SRPS EN 149:2007 [24] and SRPS EN 143:2007 [27]. Also, in the laboratory conditions, tests with NaCl aerosol are more convenient than high risk experiments with real contaminants. Our decision to take NaCl aerosol as a contaminant simulator is in line with international investigations of possible bioagents test simulators [28, 29].

Testing of the total inward leakage (TIL)

The method and the apparatus for testing in accordance with SRPS EN 149:2007 were used [24]:

- 10 smoothly shaved people were engaged in the study (without beards and sideburns) with typical user face characteristics (no significant anomalies); they have been already trained for use of identical or similar devices for respiratory protection, and selected according to specific criteria of the anthropometric matrix for the local population as defined in SORS 8746/05 [30],
- 10 samples of epidemiological masks of all models have been used (5 in the received state and 5 after temperature conditioning),
- spray-type Collison polydispersed solid aerosol from a 2% aqueous solution of NaCl was used, concentration $C_0=11 \text{ mg/m}^3$ was successfully generated, with the following characteristics: particle diameter $d_p=0.02 - 2.0 \mu\text{m}$, particle median by weight $\text{MMD} = 0.60 \mu\text{m}$, particle median per number $\text{NMD} = 0.03 \mu\text{m}$, geometric deviation $\sigma_g = 2.53$; generated particles geometric size distribution was determined by an EAA-3030 electrical particles analyzer (manufacturer TSI, USA),
- testing was performed at a test aerosol flow (TAF) of $100 \text{ dm}^3/\text{min}$.
- the concentration of the test aerosol is measured between the mask and the respondents face and in the test chamber using a Type 1100 Sodium Flame Photometer (manufacturer Moores (Wallisdown) LTD., UK).

Testing of the leakage through the filtering material (FE)

The method and the apparatus for testing in accordance with SRPS EN 149:2007 [24], shown in SRPS EN 143:2007, were used [27]:

- a required number of individually wrapped epidemiological masks (12 samples of all models) was prepared for testing: 3 samples in the state received, 3 samples were exposed to temperature conditioning, 3 samples were exposed to a procedure of simulated wear and 3 samples were exposed to tests of mechanical strength,
- spray-type Collison polydispersed solid aerosol from a 1% aqueous solution of NaCl was used, concentration $C_0=8 \text{ mg/m}^3$ was successfully generated, with the following characteristics: particle diameter $d_p=0.02 - 2.0 \mu\text{m}$, particle median by weight $\text{MMD} = 0.40 \mu\text{m}$, particle median per number $\text{NMD}=0.03 \mu\text{m}$, geometric deviation $\sigma_g = 2.53$; generated particles geometric size distribution was determined by an EAA-3030 electrical particles analyzer (manufacturer TSI, USA),

- testing was performed at a TAF of $95 \text{ dm}^3/\text{min}$,
- concentration of test aerosol was measured before and after the tested filter by a Type 1100 Sodium Flame Photometer (manufacturer Moores (Wallisdown) LTD., UK).

Testing of the resistance during inhalation (IR)

The method and the apparatus for testing in accordance with SRPS EN 149:2007 were used [24]:

- a required number of individually wrapped EMA-1 and EMN-3 masks (9 samples of both models) was prepared for testing as follows: 3 samples in the state received, 3 samples were exposed to temperature conditioning, 3 samples were exposed to a procedure of simulated wear,
- a required number of individually wrapped EMA-2 masks (12 samples) was prepared for testing as follows: 3 samples in the state received, 3 samples were exposed to temperature conditioning, 3 samples were exposed to a procedure of simulated wear and 3 samples were exposed to conditioning the flow,
- samples of all three models were tested at a continuous air flow (AF) of $30 \text{ dm}^3/\text{min}$ which was corrected to 23°C and an absolute pressure of 1 bar,
- IR of the tested samples was measured by a differential "U" water meter pressure drop. One of the excerpts was connected with the space beneath the mask at the level of the Sheffield head model mouth opening, and the other open to atmosphere.

Results

The normal statistical distribution of the obtained results was tested by the Kolmogorov-Smirnov test. It was found that the results of all tests are subject to normal statistical distribution. The summary of the results is given in Table 1 and presented as a mean value \pm SD.

The mean values of the results for total inward leakage (TIL), filtration efficiency (FE) and resistance during inhalation (IR) tests done on the epidemiological mask samples.

Table 1.

Model	TIL (%), *TAF $100 \text{ dm}^3/\text{min}$	FE (%), *TAF $95 \text{ dm}^3/\text{min}$	IR (Pa), **AF $30 \text{ dm}^3/\text{min}$
EMA-1	1.919 ± 0.007	98.810 ± 0.273	51.28 ± 1.66
EMA-2	1.903 ± 0.028	98.443 ± 0.103	51.32 ± 1.70
EMN-3	0.905 ± 0.042	99.128 ± 0.182	98.01 ± 1.79
*TAF - test aerosol flow; **AF - air flow			

According to their FE, the highest TIL and the highest IR, which are defined by SRPS EN 149:2007 standard, filtering half masks for protection against particles can be classified into three classes: FFP1 (TIL $\leq 22\%$, FE $\geq 80\%$ and IR $\leq 50 \text{ Pa}$), FFP2 (TIL $\leq 8\%$, FE $\geq 94\%$ and IR $\leq 70 \text{ Pa}$), FFP3 (TIL $\leq 2\%$, FE $\geq 99\%$ and IR $\leq 100 \text{ Pa}$). The protection offered by FFP2 or FFP3 devices includes those provided by an adequate respiratory device of a lower class [24].

The TIL results of all tested samples are very homogeneous and reproducible (standard deviation is very low). The mean value of TIL for the samples model EMA-1 is 1.919%, the model EMA-2 is 1.903% and the model EMN-3 is 0.905%, which is slightly above the limit level according to the mentioned standard. The samples of all three models thus belong to the FFP3 class.

The results of FE of all three epidemiological mask

models are also very high and very homogeneous (standard deviation is also low). The mean values of FE show that the samples of the first two models meet the criteria specified for the FFP2 class (98.810% and 98.443%), but on average slightly behind for the fulfillment of the criteria for the FFP3 class defined by the standard [24]. The mean value of FE of the third model is 99.128% and it meets the criteria for the FFP3 class. It is important to point out that the measured values of some model EMA-1 samples meet the criteria for the FFP3 class.

The results of IR of all three epidemiological mask models are also very high and very homogeneous (standard deviation is low). The mean value of resistance during inhalation at an air flow of 30 dm³/min was about 51 Pa for EMA-1 and EMA-2 samples, which is significantly less than 70 Pa and 100 Pa (limits specified for FFP2 class and FFP3 class half masks by the standard [24]). In the EMN-3 samples, the mean value of resistance was higher, as expected, at about 98 Pa, but this value is also below the limit value of permissible resistance for the FFP3 class. It is important to notice that none of the individually measured resistance values exceeds the given limits.

Discussion

It is scientifically proved that physical difficulties and disruption of cognitive and working ability are possible when respiratory protective devices are used [31-33]. This is especially important if the user is carrying out specific military tasks in combat or performs everyday tasks in peacetime conditions. The Serbian army defined military a protective full mask as a formation device of personal respiratory protection in all circumstances. The usage of this device in peacetime is questionable in the presence of other effective devices such as epidemiological masks. The reason we tested different types of epidemiological masks was to find out if there was a possibility for military personnel usage.

The personal protective equipment of this type should be effective to minimize the risk of infection transmission from persons in contact, and in the opposite direction. It should also provide comfort in normal life at work and at home, enable communication and be affordable. Protection provided by the protective device of this type depends mainly on the filtering capabilities of the built-in filtration material and on the adhesion of the fitting line on the user's face.

The mechanism of protection is based on the removal of aerosols from the inhaled air by physical binding on the filter material with a high specific surface area, providing tightness to the user's face and appropriate inhalation resistance at a flow rate of contaminated air.

Filtration materials used for producing a protective half mask may be fibers of natural or synthetic raw materials. Protection efficiency, particularly micro-organisms protection (mainly bacteria and viruses), is increased by the impregnation of these materials by silver nanoparticles [34, 35], and by combining nano-filters with primary, nonimpregnated filtration materials. Used silver nanoparticles are silver ions with a positive polarity so that all oppositely charged particles are caught in their potential pit. Since aerosols containing bacteria and viruses are negatively charged, silver ions retain them [36-38], thus making filters with nano-silver extremely protective against microorganisms.

In order to discuss the obtained results, the significance of the differences between them was tested by the Student's

t-test, with a significance level of $p < 0.05$. This is presented in Table 2 for the results of TIL, in Table 3 for the results of FE and in Table 4 for the results of IR.

Testing of the total inward leakage (TIL)

The significance of the differences between the results of TIL compared by the Students *t*-test, with a significance level of $p < 0.05$

Table 2.

Compared models	Critical <i>t</i> value	Obtained <i>t</i> value	Sig. (2-tailed)
1-2	2.2282	1.719	0.116
1-3	2.2621	75.129	0.000
2-3	2.1009	62.557	0.000

There is a significant statistical difference between the TIL results of the models EMA-1 and EMA-3. The same case is between the TIL results of the models EMA-2 and EMN-3. This fact and the smallest TIL value of the model EMN-3 (Table 1), clearly show that the use of filtration material impregnated with silver nanoparticles significantly affects the reduction of TIL.

When we observe the standard deviation of the models test results (Table 1), it can be noticed that the results of the models EMA-2 and EMN-3 have less uniformity. This is due to the imparity of hand-made prototype models. Throughout the testing, it was possible to notice the critical spots that affect the TIL in the least favourable way. Critical spots are areas where the metal bar is attached to the user's nose and the area which covers the user's chin. The metal strip on the submitted samples of half masks is located approximately 12 mm from the outer edge of the half mask, and should be no more than 7 mm in order to fit the user's nose more efficiently and in order to decrease TIL as much as possible. The construction of the half mask in the chin area is too shallow, so it negatively affects the value of the protection factor.

In spite of the noticed deficiencies, however, a low mean TIL value of all models indicates that the samples of all three models of epidemiological masks have a very good fitting on the user's face, as well.

Our results show that all three epidemiological mask models offer very reliable protection against bioaerosols in the environment and that there is a room for improvement of every model.

Testing of the leakage through filtering material (FE)

The significance of the differences between the results of FE compared by the Students *t*-test, with a significance level of $p < 0.05$

Table 3.

Compared models	Critical <i>t</i> value	Obtained <i>t</i> value	Sig. (2-tailed)
1-2	2.1448	4.353	0.001
1-3	2.0739	3.360	0.003
2-3	2.1098	11.337	0.000

There is a significant statistical difference between the FE results of all epidemiological mask models, most evident in the comparison of EMA-2 and EMN-3 models (Table 3). This fact and a higher FE value of the EMN-3 model (Table 1), confirm that silver nanoparticles dispersed in the filtration material contribute to the increased filtration efficiency of the basic epidemiological mask,

which is set as an initial hypothesis in the development of EMN-3. The fact that this model has more filter material layers than the first two ones (five layers vs. four layers) also contributes to higher FE values of the EMN-3 model.

It is important to point out that the obtained results show that the filtering material of all three models has a very high level of filtration efficiency against biological aerosols, which confirms the production quality of epidemiological masks. This confirmation is in line with the results of international investigations of levels of respirator filtration performance against nanoparticles [39, 40].

Testing of the resistance during inhalation (IR)

The significance of the differences between the results of IR compared by the Students *t*-test, with a significance level of $p < 0.05$

Table 4.

Compared models	Critical t value	Obtained t value	Sig. (2-tailed)
1-2	2.0930	0.052	0.959
1-3	2.1199	57.534	0.000
2-3	2.0930	60.910	0.000

There is a significant statistical difference between the IR results of the EMA-1 and EMN-3 models, as well as of the EMA-2 and EMN-3 models of the epidemiological mask (Table 4).

This fact and the highest IR value of the EMN-3 model (Table 1) indicates that the usage of the filtration material impregnated with silver nanoparticles greatly affects the increase of IR, almost doubling it. This results in reducing the physiological comfort, which is a subjective perception of respondents.

There is no significant statistical difference between the IR results of the EMA-1 and EMA-2 models, which indicates that the built-in exhalation valve does not affect the value of inhalation resistance. It facilitates the exhalation of carbon dioxide and water vapour, making breathing with a half mask generally easier, when in the protective position. Thus, the exhalation valve improves comfort for the user when wearing a half mask, which is also a subjective perception of respondents.

All three models of the epidemiological mask satisfy the conditions of physiological suitability specified by the standard and have a positive assessment of the physiological convenience of the epidemiological mask in terms of actual use in biologically contaminated environment.

Conclusion

The epidemiological mask has a high efficiency, physiological suitability and comfort as a protection device in the case of exposure to contamination by bioagents. The testing has shown that there are potentials for structural improvements of all tested models. The identified deficiencies of the two prototype epidemiological mask models, incurred during the mask construction and manufacturing, can be eliminated by precise machine production. Very important is the position of the metal strips which serve for mask fixation: the top edge one over the nose and the lower edge one under the chin. These two critical spots can be coated with a thin layer of sponge, as it is done by quality manufacturers, for increased wearing comfort and better protection.

The incorporated filtration material with dispersed silver nanoparticles in the EMN-3 model contributes to the increased FE and IR, and reduced TIL. According to its characteristics (high filtration material FE, low TIL and IR, low price, fast manufacturing) the most appropriate mask is the EMA-1 model - four-layered epidemiological antimicrobial mask without an exhalation valve.

Although the formational protective mask M3 has a much higher filtration efficiency of inhaled air, the epidemiological mask is more efficient in terms of user's protection and environment protection. Unlike the M3, the epidemiological mask also performs the filtration of the exhaled air, preventing the infection from spreading in the environment.

The fact that the epidemiological mask provides efficient microbial protection to and from the user and a lower resistance during inhalation makes it very operational for military personnel and physiologically more appropriate than formation devices, especially in peacetime. In accordance with the above-mentioned, it is recommended for use in peacetime together with the existing formation masks.

Acknowledgements

The Ministry of Education, Science and Technological Development of the Republic of Serbia supported this work, Grant № TR34034 (2011-2014).

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Received: 29.09.2014.

Efikasnost sredstava respiratorne antimikrobne zaštite

Individualna zaštita respiratornog trakta predstavlja ključni faktor u sprečavanju/prekidanju potencijalne epidemije/pandemije nastale prirodnim putem ili upotrebom bioloških agenasa u ratnim ili terorističkim dejstvima. Poboljšanje postojećih ili uvođenje novih pogodnijih ličnih zaštitnih sredstava u vojnu sredinu bilo bi od neprocenjivog značaja. U cilju sticanja potvrde pouzdanosti i fiziološke pogodnosti za upotrebu u zaštiti vojnog personala u slučaju objavljene epidemije/pandemije, izvršeno je ispitivanje efikasnosti zaštite i fiziološke podobnosti tri modela epidemiološke maske u uslovima simulirane biološke kontaminacije vazduha, u skladu sa standardom SRPS EN 149:2007. Na osnovu rezultata ispitivanja i njihove statističke obrade utvrđeno je da epidemiološka maska kao zaštitno sredstvo poseduje visoku efikasnost i fiziološku podobnost u slučaju kontaminacije vazduha aerosolima bioloških agenasa. Predlaže se za upotrebu u mirnodopskim uslovima ujedno sa postojećim formacijskim maskama.

Ključne reči: zaštitna maska, lična zaštita, epidemiološka zaštita, respiratorna zaštita, antimikrobna zaštita, epidemija, bioaerosol, nanočestice srebra, rezultati ispitivanja.

Эффективность средств респираторной антимикробной защиты

Средства индивидуальной защиты дыхательных путей являются ключевым фактором в предотвращении/ прерывании потенциальных эпидемий/пандемий, вызванных естественным путём или с использованием биологических агентов в состоянии войны или террористической деятельности. Усовершенствование существующих или введение новых более подходящих средств индивидуальной защиты в военную среду будет иметь неопределимое значение. Для того чтобы получить подтверждение - сертификат надёжности и физиологической пригодности для использования в защите военнослужащих, в случае опубликованной эпидемии/пандемии, было проведено испытание эффективности защиты и физиологической пригодности трёх моделей эпидемиологических масок в условиях имитирующего биологического загрязнения воздуха, в соответствии со стандартом SRPS EN 149:2007. На основании результатов испытаний и их статистической обработки было обнаружено, что эпидемиологические маски в качестве защитного агента обладают высокой эффективностью и физиологическим сходством в случае загрязнения воздуха с аэрозолями биологических агентов. Предлагается для использования в мирное время, а также вместе с существующими масками формирования.

Ключевые слова: защитная маска, индивидуальная защита, эпидемиологическая защита, защита дыхательных путей, антимикробная защита, эпидемия, биоаэрозоль, серебряные наночастицы, результаты испытаний.

L'efficacité des appareils de protection antimicrobienne respiratoires

La protection individuelle du tractus respiratoire est le facteur clé dans la prévention/interruption de l'épidémie/pandémie potentielle causée naturellement ou par l'utilisation d'agents biologiques dans les activités guerrières ou terroristes. Il serait inestimable l'amélioration des équipements de protection individuelle existants ou l'introduction de nouveaux équipements de protection individuelle plus appropriés dans l'environnement militaire. Dans le but d'acquiescer un certificat de fiabilité et de pertinence physiologique pour l'utilisation dans la protection du personnel militaire en cas de l'épidémie / pandémie publiée, on a mené une enquête sur l'efficacité de protection et de pertinence physiologique de trois modèles de masque épidémiologique dans les conditions de la contamination biologique simulée de l'air conformément à la norme SRPS EN 149: 2007. Sur la base des résultats de test et de leur traitement statistique on constate que le masque épidémiologique comme agent de protection possède une grande efficacité et une pertinence physiologique dans les cas d'une contamination de l'air par les aérosols d'agents biologiques. On le propose pour l'utilisation en temps de paix aussi avec les masques de formation existants.

Mots clés: masque de protection, protection individuelle, protection épidémiologique, protection respiratoire, protection antimicrobienne, épidémie, aérosols bio, particules nano d'argent, résultats du test.