EFFICIENCY OF SURFACE DISINFECTION BY NEBULIZATION USING CUBE ATOMIZERS OR BY USING A UV-LAMP IN A VETERINARY UNIT – PRELIMINARY STUDY

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Abstract

Maintaining proper asepsis and hygiene conditions in spaces intended for veterinary procedures remains a paramount for compliance with professional ethics. Environmental surfaces are a transitory site for microorganism accumulation and can contribute to the spread of cross-infection. The aim of the current study was to evaluate and measure the efficacy of a new nebulization technique method for surface disinfection compared to the method of disinfection with an UV lamp. The procedure was carried out in enclosed spaces used for veterinary procedures within veterinary clinics. Disinfection was performed out either with an UV-lamp or using the Cube Atomizers nebulizer, which has a revolutionary spraying system that transforms the biocide substance into microparticles, ensuring decontamination of the treated volume (air and all types of surfaces). The device ensured a successful disinfection of spaces, eliminating bacteria, and other biological pathogens. The microbiological tests were carried out before and after disinfection in both cases, on different growth mediums (Agar for the total bacteria count, Chapmann for Staphylococcus, Holmes for Streptococcus, Levine for Gram-negative Cocci and Sabouraud for fungi). An increased efficiency of disinfection was observed after using the Cube Atomizers machine, with a significant decrease in total bacteria count of almost 90-95% and the value of colony-forming units reaching 0 after nebulization in some cases; for Staphylococcus (Chapmann) there was a significant decrease, between 80-92%; for Streptococcus (Holmes) the decrease was almost 87-97%; for Gram-negative Cocci (Levine) the decrease was almost 90-99%; and for fungi (Sabouraud) the decrease was around 50%. In the case of UV-lamp disinfection, the efficiency was 30-40% lower.

Key words*: disinfection, Cube Atomizer, veterinary unit, nebulization, UV-lamp*

INTRODUCTION

Maintaining appropriate aseptic and hygienic conditions in premises intended for veterinary medical operations remains a priority in the desire for success and respect for professional ethics. The aim of the research was to assess the most vulnerable spaces in veterinary clinics, and then, applying two different disinfection methods in these spaces and measuring their effectiveness by collecting sanitation samples. Although data on nosocomial infections in veterinary medicine are limited, they are present and lately their frequency is increasing. (Ruple-Czerniak et. Al, 2013, Ruple-Czerniak et al. 2014, Stull et al., 2015) In veterinary clinics, the fact that a large proportion of the pathogens involved in causing nosocomial infections are zoonotic is a big problem for both humans and animals, considering that many zoonoses can have a serious, sometimes fatal, course. (Bîrţoiu A. et al., 2004, Gonciarov Magda 2014, Igna C., 2001, Savu et al., 2000) Examples of these are the

following: Escherichia coli, Leptospira, Brucella, Campylobacter, Salmonella, Pasteurella, Staphylococcus, Streptococcus, Clostridium, Coxiella burnetii, Chlamydia etc.(Cenariu et al., 2020, Groza I. et al.,2004, Răpuntean S. et al.,2017) All of these have particularly serious implications on organisms, and can cause genital, urinary, or mammary gland infections, gastric, skin, respiratory or various localised infections, and can even have generalised forms (septicaemia).(Groza I. et al., 1998, Răpuntean S et al., 2017, ,Savu et al.,2000) Unlike human medicine, in veterinary medicine no hygiene protocols are developed for veterinary clinics. In a veterinary hospital, where faeces and different types of secretions are always present, the susceptibility of harbouring pathogens is much higher and poses an increased risk for contamination with nosocomial and zoonotic diseases, so rigorous disinfection is crucial. Veterinarians and ancillary staff need to be educated in this regard and disinfection protocols need to be implemented in every veterinary medical unit. (Traverse et al., 2015)

MATERIAL AND METHOD

For this study, two methods of disinfection were used: nebulization and ultraviolet radiation; in closed spaces used for veterinary medical operations, in veterinary clinics in Romania, Cluj county. There was a time gap of one month between the two disinfection methods. For the nebulization method, the Cube Atomizers (distributed by the manufacturer company located in Bucharest, Modestiei Street, number 120) was used, which is equipped with a revolutionary spraying system, ensuring total decontamination of the treated volume in just a few minutes. Thus, using an authorised biocide, the machine successfully disinfects premises and all surfaces, eliminating bacteria, fungi, viruses and other biological pathogens. Its revolutionary system breaks down the biocide into micro-particles, which remain in the air for a long time without leaving residues. For the ultraviolet disinfection method, a UV-C ultraviolet light lamp was used (distributed by the manufacturing company located in Iasi, Bulevardul Chimiei, number 12). Compared to the radiation method, where the time of use is long, the Cube fogging machine ensures disinfection of the entire surface in just a few minutes and is very easy to use. Unlike the UV-C disinfection method, the Cube nebuliser guarantees safety for the user, the environment and all treated materials. Ultraviolet lamps require a longer period of time to disinfect the space and can cause skin and eye damage and over time they cause degradation of treated surfaces, and by generating ozone can cause respiratory tract diseases (Buonanno M. et al.,2017, FDA, 2021). In order to apply the methods proposed for the study of the research topic, sanitation samples were taken both before the start of operations (Before test) and after disinfection (After test).

A. Materials needed

In the application of both disinfection methods, the following materials were required for the sampling and the study (Fig. 1): Non-sterile disposable gloves, protective equipment (disposable gown), protective goggles, cleaning materials(detergent and wipes), disinfectant, waste collector, apparatus for measuring room dimensions and volume (laser telemeter), sterile swabs with culture medium, sterile test tubes specially prepared for sampling, petri dishes and other utensils necessary for sampling for bacteriological analysis, a power supply (earthed socket) and a timer. In addition, for the nebulization method we had the Cube Atomizers nebulization apparatus, measuring utensils for preparing the quantities of the necessary disinfectant product and the Biosan Steridet disinfectant solution. This is a pink powder with strong disinfectant action, containing Pentapotassium Bis (peroxymonosulphate) Bis (sulphate) as active substance. For the radiation

disinfection method we also required the UV-C lamp.

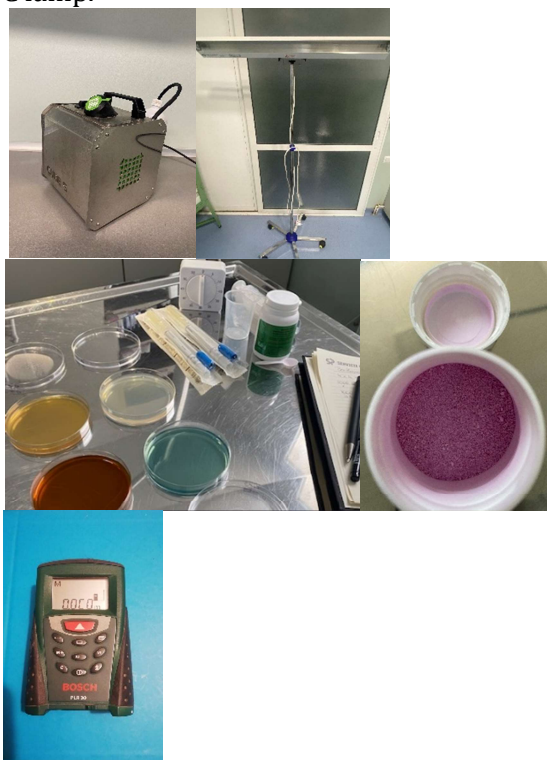


Fig. 1 - Materials needed: Cube Atomizers, UV-C lamp, Petri dishes, Sterile swabs with culture medium, Biosan Steridet disinfectant solution, Timer, Graduated beakers, Laser telemeter (Original)

B. Preparing the space for disinfection and actual nebulization:

Mechanical cleaning according to general cleaning protocols was carried out prior to the start of nebulization. After preparation of the space, the volume of the room was calculated using the laser range finder to determine the time required to operate the machine and the amount of disinfectant material required for use. Finally, the space was closed as tightly as possible. Protective equipment was donned and the machine was prepared and supplied with disinfectant. Then, using a grounded socket, the Cube Atomizers were placed in a corner of the room, with the nozzle oriented diagonally across the room. (Fig.2) The windows and doors in the room were closed, it was ensured that no other person was in the space and access was restricted to any person until the space was ventilated after the cycle was completed (nebulization/activation/aeration). The button was placed in the ON position and the room was left, closing the door. The appliance was left to operate and waited until nebulisation was complete (completion of the process is identified by the cessation of the specific sound made by the appliance when operating). The switch was set to the OFF position, the machine was removed from the room and the room was kept closed for the contact time and then the room was ventilated. After use, the apparatus is wiped with a cloth and stored in a place protected from the weather and out of direct sunlight.

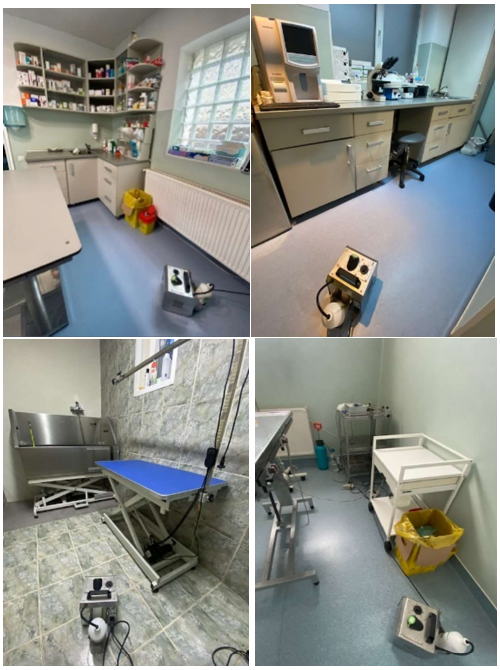


Fig. 2 - Positioning and preparing the nebulization machine (Original)

C. Preparing the space for disinfection with the UV-C lamp

Prior to the start of disinfection of the space using the UV-C method, mechanical cleaning of the space and surfaces was carried out according to general cleaning protocols. After preparation of the space, protective equipment was donned, the UV-C lamp was installed, positioned in the centre of the room, connected to a grounded outlet and left to operate for 30-60 minutes, depending on the

surface area of the space, according to the specifications provided by the manufacturer. (Fig.3) During the use of the UV-C lamp, the space was closed and the perimeter secured so that no one could enter. After the end of the exposure time, the UV-C lamp was closed and wiped with a cloth.

Fig. 3 – Positioning and preparing the UV-C lamp (Original)

D. Sample collection and analysis

Sterile swabs with culture medium were used to collect sanitation samples for the determination of microorganisms. Both before and after the premises were disinfected, samples were collected with these swabs from various surfaces in the room (tables, floors, surgical lights, inhalation anaesthesia machines, cabinets and other equipments) (Fig. 4). Samples collected in this way were seeded onto solid culture media in Petri dishes. The plates were then incubated and after incubation the developed colonies were counted. Simple agarose (nutrient agar) known as agar is used to determine the total number of germs. By frequency in air and on all surfaces, as well as implications for pathology, in addition to the total number of germs, the following pathological agents were determined in particular: staphylococci on Chapmann medium, streptococci on blood agar, sodium azide and crystal violet (Holmes medium), gram-negative germs on Levine medium, and fungi on Sabouraud medium.

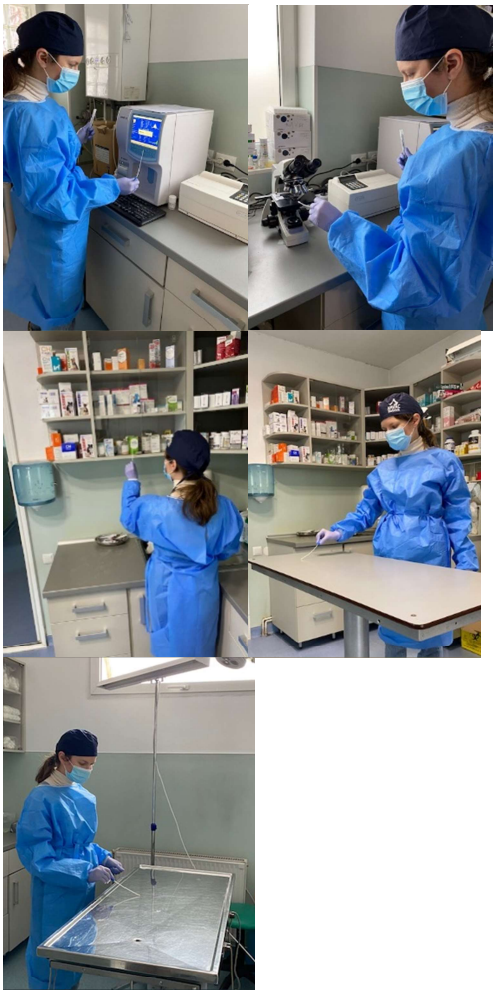


Fig. 4 – Sampling procedure (Original)

RESULTS AND DISCUSSIONS

The results of the sanitation tests before the start of disinfection procedures (Before test) and after mechanical cleaning and disinfection (After test) in the premises for veterinary medical procedures in veterinary clinics can be seen in the tables (Table 1 and Table 2) and graphs (Graph 1 and Graph 2) below:

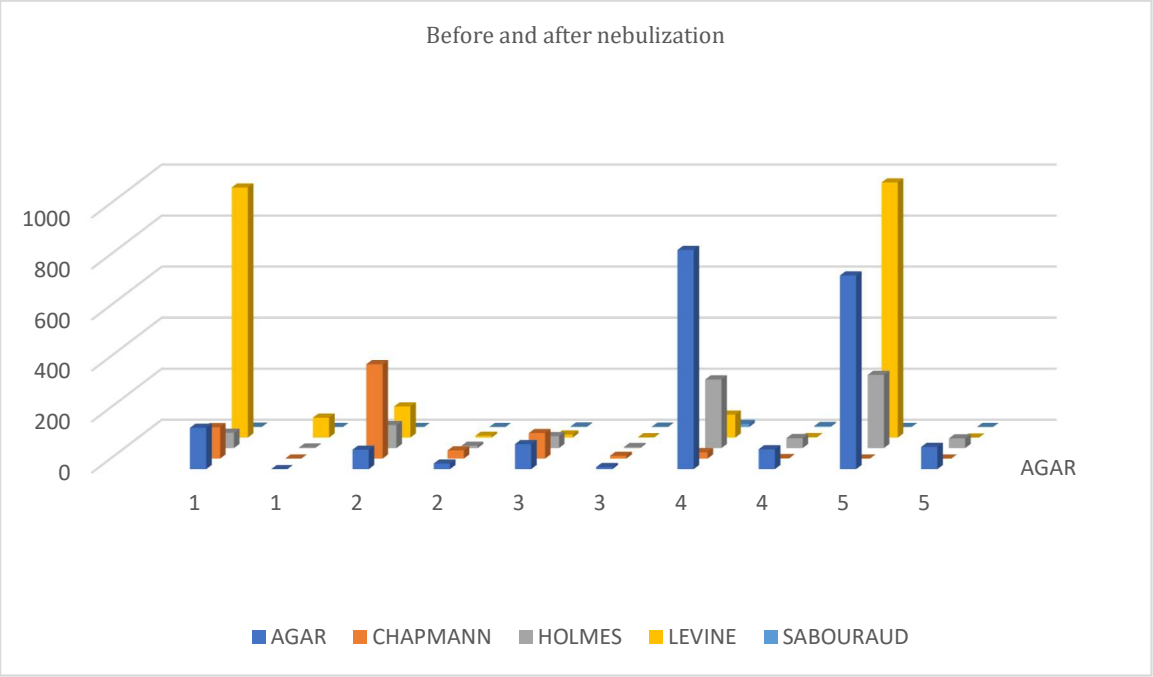
Table 1 - Results of sanitation tests before and after nebulization

SAMPLE	**AGAR (Total bacteria count) (ufc/m^3)**	**CHAPMANN (Staphylococcus) (ufc/m^3)**	**HOLMES (Streptococcus) (ufc/m^3)**	**LEVINE (Gram-negative Cocci) (ufc/m^3)**	**SABOURAUD (fungi) (ufc/m^3)**
1 Before disinfection	162	123	60	980	1
1 After disinfection	0	0	2	78	0
2 Before disinfection	76	370	90	122	0
2 After disinfection	22	32	9	7	0
3 Before disinfection	98	100	47	12	2
3 After disinfection	8	11	4	1	1
4 Before disinfection	859	25	269	90	12
4 After disinfection	78	2	39	2	4
5 Before disinfection	759	0	287	1021	0
5 After disinfection	87	0	38	0	0

Table 2 - Sanitation test results before and after using the UV-C lamp

SAMPLE	**AGAR (Total bacteria count) (ufc/m^3)**	**CHAPMANN (Staphylococcus) (ufc/m^3)**	**HOLMES (Streptococcus) (ufc/m^3)**	**LEVINE (Gram-negative Cocci) (ufc/m^3)**	**SABOURAUD (fungi) (ufc/m^3)**
1 Before disinfection	380	180	43	879	3
1 After disinfection	190	107	23	410	3
2 Before disinfection	58	870	78	129	1
2 After disinfection	23	407	47	70	1
3 Before disinfection	147	120	73	21	1
3 After disinfection	67	69	37	9	1
4 Before disinfection	127	40	79	82	3
4 After disinfection	43	17	38	50	3
5 Before disinfection	175	1	85	53	1
5 After disinfection	63	1	34	27	1

Graph 1 - Sanitation sample results before and after nebulization



Graph 2 – Sanitation sample results before and after using a UV-C lamp

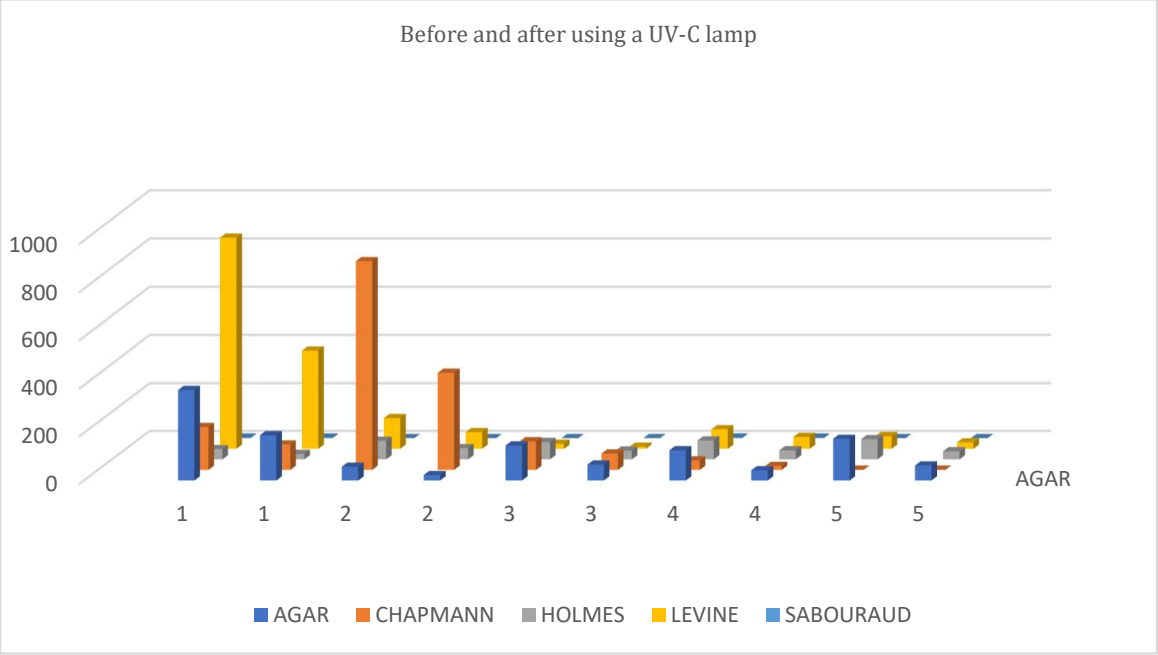


Fig. 5 - Sanitation samples (Culture medium swabs) (Original)

After keeping the samples in the thermostat at the appropriate temperature and conditions, the results were interpreted. For the nebulisation method used, in the samples cultivated on Agar culture medium, a significant decrease in the total number of germs was observed with an average of 90-95% after disinfection, in some case the success rate was 100%, reaching a value of 0 after disinfection. When using Chapmann medium in some case we did not find the presence of staphylococci either at the beginning of the experiment or after the application of disinfection, but in the cases where we found the presence of staphylococci, the number of staphylococci was reduced to 80-92% after disinfection and in some cases the value reached 0 (100%) after disinfection. In the case of Holmes culture media, the number of streptococci was observed to decrease by an average of 87-97% following disinfection. The same was true for the Levine medium, specific for the isolation of gram negative bacteria, with a decrease of 90-99% on average. In the case of Sabouraud medium a decrease of almost 50% on average was observed. In the case of using the UV-C lamp, the success rates were in average: 57,8% for the total number of germs, 48% for staphylococcus, 49% for streptococcus, 48% for gram-negative cocci, and for fungi there was no change, the results were the same after disinfection like they were before disinfection. Thus, we found a 30-40% higher efficacy of the nebulisation machine used in this study than in the case of using the ultraviolet lamp as a disinfection method.

CONCLUSIONS

Following the application of disinfection methods in veterinary medical premises the following were observed:

-in the case of using the Cube Atomizer nebulizer, in most of the germ categories, in many of the cases, the value of the number of colony-forming units reached 0 after nebulization (the efficiency was 90-97% for the total number of germs, 85-95% for Staphylococci, 90-99% for streptococci, 92-99% for gram-negative bacteria and around 50% for fungi) which is of particular importance considering the obligation of a high degree of hygiene in these premises;

-efficiency is 30-40% higher for the nebulisation method proposed for this study than for the UV-C radiation method;

-the Cube Atomizer is even more efficient as it does not require a long time to use, unlike the UV-C lamp;

-the nebuliser leaves no residue, uses a small amount of disinfectant, is easy to handle and does not require the unit to change the disinfectant it prefers;

-the small amount of disinfectant used for the nebulization machine and the short application time leads to lower costs and a short payback of the initial investment, unlike the UV lamp which due to the long use time requires frequent change of the UV-C tube, thus leading to additional costs;

-the nebulization method used has the advantage of being able to disinfect surfaces (floors, ceilings, appliances, windows, furniture, etc.) on all sides, including the back, areas underneath, edges, pipes, etc.), which would take much longer or be impossible to

achieve with the UV-C lamp disinfection method, and would result in partial disinfection of surfaces compared to the above;

-unlike the UV-C disinfection method, the Cube nebuliser guarantees safety for the user, the environment and all treated materials. The ultraviolet lamp requires a longer period of time to disinfect the space and can cause skin and eye damage if not handled carefully and can cause irritation to the respiratory tract by generating ozone. If used for a long time, the lamp causes degradation of treated surfaces;

-periodic disinfection, based on a planning schedule, with Cube Atomizers in areas used for veterinary medical operations can ensure a high level of hygiene at all times and prevent nosocomial infections;

-it is necessary to educate veterinary staff about the obligation to maintain a high level of hygiene in veterinary clinics;

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