

RESULTS OF FUNGICIDE ACTIVITY OF CID 20 DECONTAMINANT ON ASPERGILLUS GENE FUNGI (ATCC 16404 - *ASPERGILLUS BRASILIENSIS*)

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Abstract

The objective of this paper was to determine the fungicidal activity of the CID 20 decontaminant on a strain of Aspergillus brasiliensis, ATCC 16404.

In order to accomplish this goal, the following work plan was drawn up: an artificial contamination of 100 cm² surfaces was made; 1 strain of Aspergillus brasiliensis was used to prepare a fungal suspension with a 1 McFarland density using a Biosan density.

From the fungal suspension with 1 McFarland density, several work solutions with different concentrations were made: 0.25%, 0.5%, 1%.

Several surfaces were contaminated with the suspension of Aspergillus brasiliensis, then these surfaces were sprayed with decontaminant CID-20, and finally, sanitation tests were taken to verify the effectiveness of the decontamination with this product.

The conclusion of our research was that decontaminant CID 20 has effective action on Aspergillus genus.

Key word: decontaminants, fungal suspension, Aspergillus brasiliensis.

INTRODUCTION

Decontamination is a key element in bird breeding, which is why the disinfectant substances in broiler chickens are very carefully chosen to get the most effective results.

The location of thousands of birds in a closed, warm and dusty environment is very favorable for transmission of contagious diseases (Collins, 2007).

In addition, the selection of birds for faster growth and increased meat production has left the bird's immune system less able to cope with infections and there is a high degree of genetic heterogeneity in the population, which makes the spread of the disease easier (Delany, 2003).

Microbiological contamination can be prevented and controlled using appropriate managerial practices and health products such as disinfectants (MSU, 2008).

Space decontamination units (such as C-ultrasound systems and hydrogen peroxide) help reduce environmental contamination after cleaning and disinfection (Rutala A. et al., 2013).

In the atmospheric air there is always a varied microbial flora, whose number varies according to the place and time of harvest, having higher values near the soil than at altitude (Popescu D. et al., 1983).

Biosecurity measures in broiler breeding farms can be grouped into two categories:

- prevention measures that address the potential risk factors on the farm (biotic and abiotic);
- organizational measures to prevent infectious and parasitic agents from entering the farm (Vacaru-Opris I, 2004).

By reducing the number of pathogens, the potential for disease in the poultry farm is reduced (Block, 2001), which will result in a good slaughter yield - the commercial yield value for broiler chickens must have limits between 80% and 84%, with an average of 82% (Garlic MG, 2008).

When using on farms, not all disinfectants show the work we can expect in standard tests (HACCP Manual, 2008), it is precisely for this reason that the effectiveness of the decontaminants we use for the disinfection of poultry breeding sites should be checked.

In order to prevent the introduction of pathogenic germs, disinfection of all means of transport and human beings is applied in zootechnical units within the sanitary filter. In the veterinary units and objectives, prophylactic disinfection should be applied daily, using in particular chemical means, but also some physical means such as heat, ultraviolet radiation, etc. (Decun M., 1981).

CID 20 is one of the decontaminants used in poultry establishments (Figure 1) and is based on 5 different active ingredients: quaternary ammonium compounds, aldehydes, alcohol and stabilizing agents, buffers, surfactants, agents, separators, foaming potentiators, nebulizing potentiators and corrosion inhibitors.



Fig. 1 Decontaminating substance Cid 20

The balanced formula of CID 20 determines extraordinary efficiency against bacteria, spores, viruses and fungi.

Advantages of using CID 20:

- is excellent for disinfection of animal shelters, materials, surfaces, transport equipment for birds, pigs, dairy and meat cows, incubators and processing plants;
- financially advantageous;
- has fast and long-lasting action;
- is non-corrosive;
- has a guaranteed shelf life of 3 years.

MATERIAL AND METHOD

In order to achieve this goal, the following work plan was made: Artificial contamination of 100 cm² surfaces was made using a 10 x 10 cm aluminum template (Figure 2); a strain of *Aspergillus brasiliensis* ATCC 16404 was used to prepare a fungal suspension with a density of 1 McFarland by density.

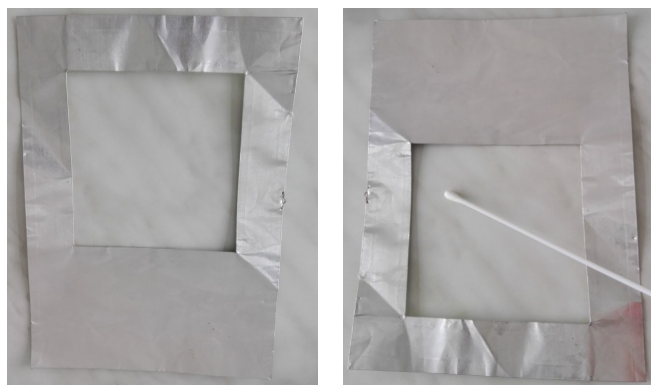


Fig.2 Aluminum template for sanitation sampling (S = 100 cm²)

The suspension was obtained in the following manner: Several colonies of *Aspergillus brasiliensis* were harvested using a disposable bacteriological dish that were placed in a distilled water tube until a 1 McFarland density (Figure 3).



Fig.3 Biosan Densimat: - Making a working solution of 1 McFarland from the fungal strain of *Aspergillus brasiliensis* ATCC 16404

The experiments were divided into several stages:

- In the first step, working solutions of CID 20 were prepared with different concentrations: 0.25%, 0.5%, 1%;
- in stage II several surfaces were contaminated with the suspension of *Aspergillus brasiliensis*;
- after the drying of the surfaces was followed the third stage, the spraying of the contaminated surfaces with the solutions of CID 20 at the three concentrations; for each concentration used, 3 contact times were used: 10 minutes, 20 minutes, and 30 minutes respectively;
- the last stage consisted of collecting the sanitation tests on the 9 surfaces and putting them into work according to SR ISO 21527-1: 2009.

Preparation of solutions of CID 20: 0.25%, 0.5%, 1%

CID 20 0,25%:

0,25 ml CID 20	+	100 ml Distilled sterile water
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CID 20 0,5%:

0,5 ml CID 20	+	100 ml Distilled sterile water
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CID 20 1%:

1 ml CID 20	+	100 ml Distilled sterile water
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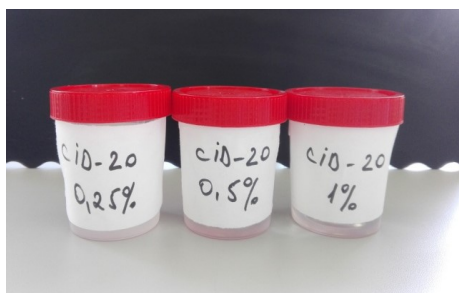


Fig.4 Solutions of CID 20 (0.25%, 0.5%, 1%)

RESULTS AND DISCUSSION

As a result of the analyzes, the result was that the CID 20 concentration of 0.25%, 0.5% and 1% had effective fungicidal action; all Dichloran Rose-Bengal Chloramphenicol Agar plates sampled with samples collected from decontaminated surfaces with this decontaminant were sterile, indicating no microbial growth (Table 1, Figure 7; 8; 9).

Table 1

Results obtained after decontamination with CID - 20

Substance used	The concentration / Contact time	Results obtained prior to contamination with A. brasiliensis	Results obtained after contamination with A. brasiliensis	Results obtained after decontamination with A. brasiliensis
CID - 20	0,25% - 10'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	0,25% - 20'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	0,25% - 30'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
CID - 20	0,5% - 10'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	0,5% - 20'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	0,5% - 30'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
CID - 20	1% - 10'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	1% - 20'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²
	1% - 30'	Absent/100 cm ²	Present/100 cm ²	Absent/100 cm ²



Fig.5 Plate with DRBC medium: before contamination with *Aspergillus brasiliensis*



Fig.6 Plate with DRBC medium: after contamination with *Aspergillus brasiliensis*

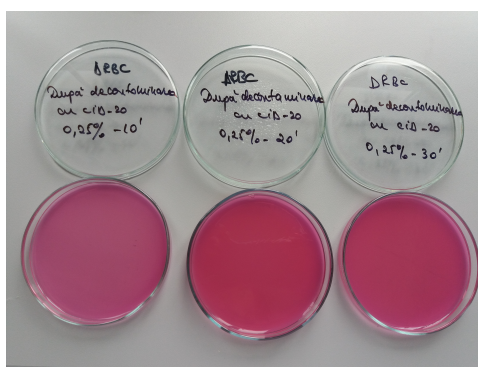


Fig.7 DRBC media plates: after decontamination with CID 20 - 0.25%

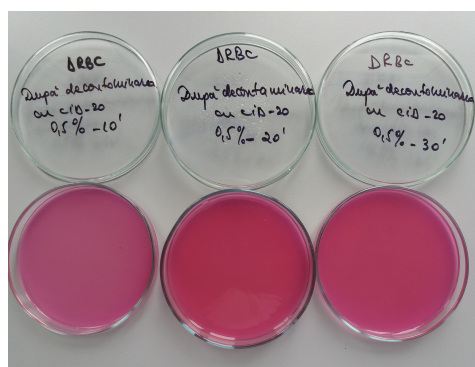


Fig.8 DRBC media plates: after decontamination with CID 20 - 0.5%

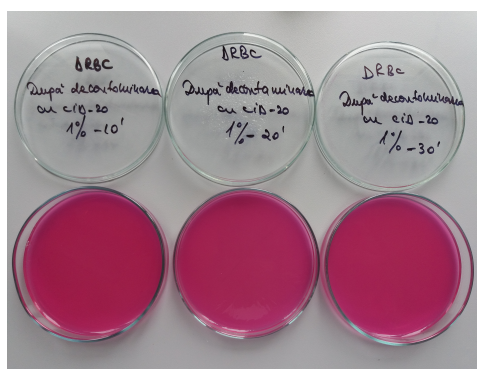


Fig.9 DRBC media plates: after decontamination with CID 20 - 1%

In 2009, Belgian researchers studied the effect of decontaminant CID 20 along with D50 and Virocid in order to see the decontamination action it has on strains of *Salmonella enteritidis*, *Escherichia coli* and *Enterococcus* spp., And to evaluate *Escherichia coli*, *Enterococcus faecalis* and *Enterococcus hirae* as potential indicator organisms by the quantitative suspension disinfection test.

Disinfection tests showed that none of the indicators studied was as sensitive or less susceptible to products than SE, although we consider that *Escherichia coli* is still a possible candidate for the following reasons: Firstly, *Escherichia coli* was more resistant than *Enterococcus* spp. ; Secondly, since *Escherichia coli* was more susceptible to disinfection than *Salmonella enteritidis*, the researchers concluded that when *Escherichia coli* is found after cleansing and disinfection, there is a major possibility that *Salmonella enteritidis* may not be completely eliminated and may be present.

However, when *Escherichia coli* was not found after cleansing and disinfection, there is still a small chance that *Salmonella enteritidis* is present (Dewaele I. et al., 2009).

In 2016, several Nigerian researchers have been studying to see if bursal disease virus in birds can be inactivated with polidin® (iodoform compound), V-ox® (inorganic peroxide compounds), CID20® (quaternary ammonium chloride, aldehydes and alcohol), terminator III (phenols) and glutasan (aldehyde and quaternary ammonium chloride).

However, the results were not satisfactory, failing to eliminate the IBD virus in birds (Aliyu H. B. et al., 2016).

Researchers from China also conducted studies in 2007 to test the effectiveness of CID 20 in laboratory and equipment lab rooms.

Their results were satisfactory: the number of bacteria on the roof, wall and corridor decreased from 88.2% to 100% after disinfection, which is why the researchers concluded that the decontaminant Cid 20 can be used for the

current disinfection of the spaces and equipment for laboratory animals (DU Xiao-yan et al., 2007).

CONCLUSIONS

Analyzing the results, we can conclude that the decontaminant CID-20 with a concentration of 0.25%, 0.5% and 1% had effective action on *Aspergillus* genus, and can be used with confidence for the decontamination of livestock .

The fact that the product is effective at a low concentration (0.25%) means lower costs with decontamination, which is economically beneficial to users.

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