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EVALUATION OF FUNGAL INCIDENCE IN BROILER FARM

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Abstract

This research aimed the isolation and identification of mycoflora from forage, water, air, litter and sanitation swabs collected during the year 2011. Collecting and processing of samples was done according with the literature data and current standards. Therefore the air samples were collected and processed by method of sedimentation and the forage, water, litter and sanitation swabs samples have been harvested and processed according to ANSVSA Order no. 25 of 19 March 2008, SR EN ISO 6887-1/2002 and ISO 7218 /2007. Through the qualitative mycological exams have been identified 11 genres (Aspergillus spp.*, Penicillium* spp.*, Mucor* spp., *Absidia* spp., *Rhyzopus* spp., *Alternaria* spp., *Ulocladium* spp., *Cladosporium* spp., *Fusarium* spp., *Candida* spp., *Rhodotorulla* spp.*) and 12 species (Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus, Aspergillus glaucus, Absidia corymbifera, Fusarium oxisporum, Rhodotorulla rubra, Candida albicans, Candida sake, Candida rugosa, Candida famata and Candida lusitaniae).*

Key words: *broiler,fungi,incidence.*

INTRODUCTION

A permanent objectiv in broiler farms is monitoring hygiene status. In parallel with hygienisation programs, for efficient fight against pathogenic microorganisms is necessary periodic evaluation of the microbial species existing in farm. The diseases caused by pathogenic fungi, like all of infectious diseases, causes the economic losses through high morbidity and mortality and decreased the production indicators. Among the fungal infection of poultry, the most frequent reported in the literature are: aspergillosis and candidosis. Aspergillosis is the most important fungal infection, being frequent caused by *Aspergillus fumigatus* and rarely by *Aspergillus flavus* or the *Aspergillus niger*. The disease mainly affects respiratory tract of broilers and develops as a bronchopneumonia (Milos C. et al, 2011; Stoenescu Virginia., 1964). Candidosis is encountered frequently in respiratory mucosa and air sacs of poultry. The pathogenic fungi (in the presence of mucosal injury) such as *Penicillium* spp.*, Mucor racemousus*, may cause especially at hens, pseudotubercles nodules and clinical signs similar to those of aspergillosis (Stoenescu Virginia., 1964). Other authors (Coman I., 1985) have shown that fungi of the Penicillium spp., *Alternaria* spp., *Aspergillus* spp., or *Fusarium* spp., get into the organism of poultry through feed contaminated with spores, thus becoming toxic for poultry to which triggers a hemorrhagic syndrome.

MATERIALS AND METHODS

From January 2011 to September 2011, a total of 50 samples were taken according to ANSVSA Order no. 25 of 19 March 2008, from commercial broiler farm. Of all 50 samples, 10 samples were represented by the feed, 10 by the water, 10 by the air, 10 by the litter and 10 by the swabs. All samples were aseptically transported to the laboratory and were stored at 4°C for fungal analyses. The air samples harvested through the Koch method (Coman I., 1997) on the surfaces of Sabouraud Chloramphenicol Agar were immediately thermostated, for 7-14 days at 25°C. After that the fungal strains develop were identified. The samples of feed, water, litter and swabs were processed according to current standards (SR EN ISO 6887-1/2002; SR EN ISO 7218 /2007). Of the inoculum resulting from the processing of samples were performed dilutions from which were apply each 0,1ml to the center of a dry plate with Sabouraud Cloramphenicol Agar and spread with loop of the entire surfaces for a

better absorption. Then plate was incubated at 25°C for 7-14 days. To obtain pure culture were transferred onto Malt Extract Czapek for the identification of isolate fungi. Most filamentous fungi can be identified based on a combination of colonial morphology and microscopic features in accordance with keys (http://www.mycology.adelaide.edu.au/Keys, http://www.doctorfungus.org,http://www.cbs.knaw.nl). Slides were prepared for identification of mycelium and macroconidia with lactophenol blue staining method. Yeasts were subcultured and identified with the Api 32 C system (BioMerieux France).

RESULTS AND DISCUSSIONS

In Table 1. is shown the incidence of fungal genres and species isolated from broilers farm.
Table 1. Fungal incidence in forage, water, air, litter and swabs

Table 1. Fungal incidence in feed, water, air, litter and swabs

	Feed No.samples/10		**Water** No. samples/10		**Air** No. samples/10		**Litter** No.samples/10		**Swab** No.samples/10		**Grand total** No.Total samples/50	
	Strains		Strains		Strains		Strains		Strains		Strains	
Genus / Species	nr.	%	nr.	%	nr	%	nr.	%	nr.	%	nr.	%
Aspergillus niger	3	30%	0	0%	3	30%	3	30%	2	20%	11	22%
Aspergillus flavus	2	20%	1	10%	2	20%	2	20%	1	10%	8	16%
Aspergillus fumigatus	2	20%	1	10%	7	70%	5	50%	5	50%	20	40%
Aspergillus glaucus	1	10%	0	0%	0	0%	2	20%	1	10%	4	8%
Penicillium	4	40%	3	30%	3	30%	7	70%	5	50%	22	44%
Mucor spp.	3	30%	0	0%	1	10%	3	30%	2	20%	9	18%
Absidia corymbifera	1	10%	0	0%	0	0%	2	20%	1	10%	4	8%
Rhyzopus spp.	0	0%	0	0%	1	10%	1	10%	1	10%	3	6%
Alternaria spp.	3	30%	1	10%	1	10%	3	30%	2	20%	10	20%
Ulocladium spp.	2	20%	0	0%	1	10%	1	10%	1	10%	5	10%
Cladosporium spp.	2	20%	1	10%	2	20%	2	20%	1	10%	8	16%
Fusarium oxisporum	3	30%	1	10%	1	10%	2	20%	1	10%	8	16%
Total molds	**26**		**8**		**22**		**33**		**23**		**112**	
Rhodotorula rubra	2	20%	2	20%	0	0%	4	40%	2	20%	10	20%
Candida albicans	3	30%	2	20%	0	0%	3	30%	1	10%	9	18%
Candida sake	0	0%	0	0%	0	0%	1	10%	0	0%	1	2%
Candida rugosa	0	0%	0	0%	0	0%	1	10%	1	10%	2	4%
Candida famata	0	0%	1	10%	0	0%	0	0%	0	0%	1	2%
Candida lusitaniae	0	0%	0	0%	0	0%	1	10%	0	0%	1	2%
Total yeasts	**5**		**5**		**0**		**10**		**4**		**24**	
Grand total strains	**31**		**13**		**22**		**43**		**27**		**136**	

Form the data analysis presented in Table 1 and Figure 1., follows that of the total samples taken been identified 136 strains of fungi out of which 112 (82,35%) were molds and 24 (17,65%) yeasts.

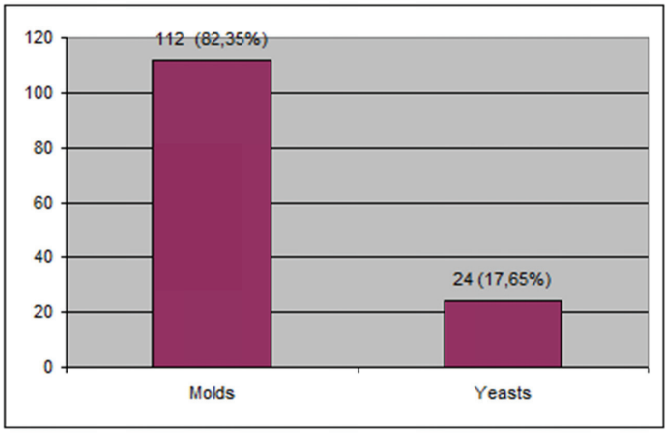

Figure 1. Incidence of molds and yeasts in broilers farm

More precisely of the 50 samples taken from the farm were found 11 strains (22%) of *Aspergillus niger*; 8 strains (16%) of *Aspergillus flavus*; 20 strains (40%) of *Aspergillus fumigatus*; 4 strains (8%) *Aspergillus glaucus*; 22 strains (44%) of *Penicillium* spp.; 9 strains (18%) of *Mucor* spp; 4strains (8%) of *Absidia corymbifera*; 3 strains (6%) of *Rhyzopus* spp; 10 strains (20%) of *Alternaria* spp; 5strains (10%) of *Ulocladium* spp; 8 strains (16%) of *Cladosporium spp*; 8 strains (16%) of *Fusarium oxisporum*; 10 strains (20%) of *Rhodotorulla rubra*; 9 strains (18%) of *Candida albicans*; 1 strain (2%) of *Candida sake*; 2 strains (4%) of *Candida rugosa*; 1 strain (2%) of *Candida famata* and 1 strain (2%) of *Candida lusitaniae*. From the data presented in Table 1, it results that samples of litter are the most intense contaminated (43 strains), being followed by the feed samples (31 strains) and the sanitation swabs (27 strains). From the feed, water and litter samples the most frequent isolated fungi species is *Penicillium* spp. (Figure 2, 3) *Aspergillus fumigatus* (Figure 4, 5) is the most fequent isolated fungi species from the air and litter samples. The yeasts *Rhodotorulla rubra* (Figure 6, 7) and *Candida albicans* (Figure 8, 9) were most frequent isolated from the litter samples.

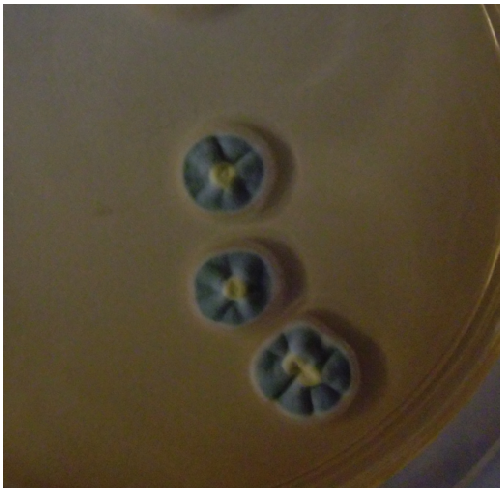

Figure 2. *Penicillium* spp. macroscopic

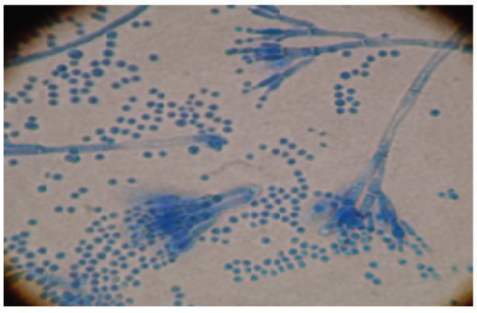

Figure 3. *Penicillium* spp. microscopic

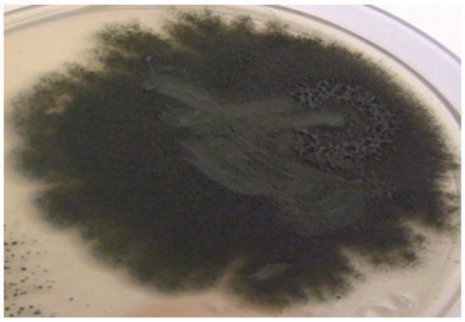

Figure 4. *Aspergillus fumigatus* macroscopic

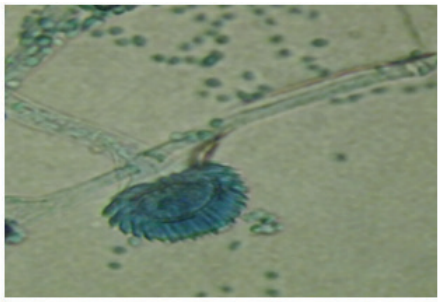

Figure 5. *Aspergillus fumigatus* microscopic

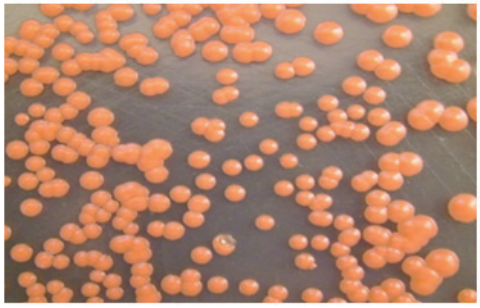

Figure 6. *Rhodotorulla rubra* macroscopic

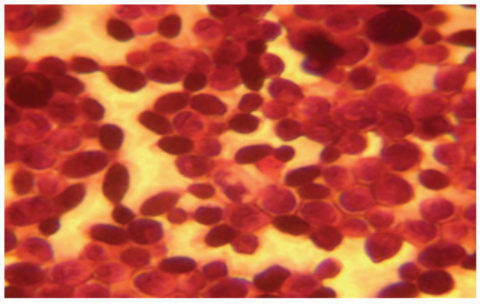

Figure 7. *Rhodotorulla rubra* microscopic

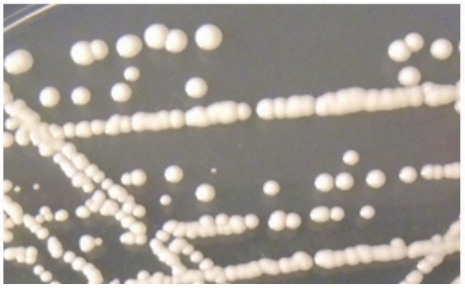

Figure 8. *Candida albicans* macroscopic

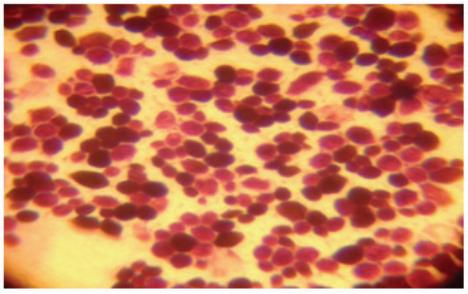

Figure 9. *Candida albicans* microscopic

Analyzing the results obtained of the mycological research about the incidence of fungi, it follows that are in accordance with the data in the literature. Therefore, Arne (Arne P. et al, 2011) show in his analysis that *Aspergillus fumigatus* grows in litter of poor quality, in feed stored in poor conditions and in the air from broilers farm. Furthermore, inadequate ventilation increases the risk of exposure of poultry to inhalation of spores causing high morbidity and mortality. The results of present study are also in agreement with the findings of Azarakhsh (Azarakhsh Y. et al, 2011) who reported a higher incidence of *Aspergillus spp* in feed.

CONCLUSIONS

The fungal flora isolated and identified from forage, water, air, litter and swabs samples was various. Also were have identified some pathogenic fungi can exert immuno-suppression on broilers.

Through the macroscopic, microscopic and biochemical tests were isolated and identified 11 genera and 12 species of molds and yeasts. Presence of opportunistic pathogens from *Aspergillus* poses a risk of invasive aspergillosis in poultry farm.

Compared to the rest of the samples, the litter have proved to be the most intensely contaminated (43 strains), the most common fungi isolated were *Penicillium* spp., *Aspergillus fumigatus, Candida albicans* and *Rhodotorulla rubra*.

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