Vet.Med.J.,Giza. Vol.43,No.2.(1995):157-166.

SEDIMENTATION RATE (A NEW EQUATION) FOR AIRBORNE FUNGI IN A LIBYAN ABATTOIR

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SUMMARY

A total of 80 air samples from different compartments of slaughter house Shahaat (A libyan abattoir) were examined mycologically. A number of 360 mould strains were recovered , at the top were dark moulds (87.22%), Aspergillus (6.67%, Penicillium (2.22%), Fusarium (1.94%). Mucor and Absidia were also isolated. Sedimentation rate of cfu of moulds on one cm^2 was calculated

INTRODUCTION

Air is the most important source of mould spores and their elements. Sedimentation of mould spores was recorded on carcases from air (Hirst, 1953; Pady, 1957; Hudson, 1969; Refai and Loot, 1969,Lowry and Gill, 1984; Mansour, 1986; Hamdy et al., 1990).

Refai and Loot (1969) examined air in slaughter houses and butcher's shops. They stated that aspergillus and Penicillium were the most prevalent isolates, while other moulds were also found as Rhizopus, Mucor, Alternaria, Cephalosporium, Scopulariopsis, Pullularia and Streptomyces.

Ahmed et al. (1984) recorded airborne fungi in different animal pens as buffaloes, cattle , sheep, rabbits and chickens,. Isolated strains were Aspergillus, Penicillium, Cladosporium, Mucor, Alternaria and Fusarium.

Baxter and Illiston (1976) recovered some airborne fungi from yards of sheep, slaughter halls and deboning rooms . The isolated strains were Cladosporium herbarum, Cladosporium Cladosporioides, Epicoccum purpurascens, Alternaria alternata, Penicillium expansum and Sportichium carnis.

Lowry and Gill (1984) stated that falling and sedimentation of airborne fungi on carcases is not uncommon. According to this fact, the air is considered as one of the primary sources of contamination of carcases with mould spores.

Mansour (1986) investigated air in Munich abattoir mycologically. The most predominent airborne fungi were Aspergillus (29.5%), Cladosporium (14.1%), and Penicillium (11.1%). Other mould genera were also recovered such as Scopulariopsis, Absidia, Mucor, Fusarium, Rhizopus, Acremonium, Geotrichum, Thaminid um, Trichoderma, Trichopohyton, Ulocladium, and Exophialia.

Hamdy et al. (1990): recovered 806 mould strains from 50 air samples of camel and cattle sluaghter halls. Aspergillus and Dematiaceous

hypohomycetes (dark moulds) constituted over 85% and 95% of air-borne fungi in the above mentioned halls respectively. Aspergillus was on the top of the airborne Fungi in the examined locations (33.88% and 51.18% respectively). Other mould strains were also isolated as Trichoderma, Rhizopus, Absidia. Aeromonium, Fusarium, Geotrichum, Scopulariopsis, Mucor and Paecilomyces.

Mansour et al. (1990) exmined also camels and catttle slaughter halls and their surroundings including air with special reference to Dematiceous hyphomycetes. Airborne dark moulds were Aspergillus niger, Cladosprium and Alternaria.

Refai et al. (1993) examined mycologically 200 air samples in modern Egyptian abattoris. The most prevalent isolates were Aspergillus (32.91%), Dematiceous moulds (26.41%) and Penicillium (20.13%). Aspergilli could be identified as A.niger, A. flavus, A. fumigatus, A. ochraceous, A. tamarii, A. terreus and A.parasiticus, Cladosporium constituted 14.84% of the total isolates, on the other hand Alternaria was 5.85%. Other moulds were also recorded as Paecilomyces, Scopulariopsis, Trichoderma, Mucor, Rhizopus, Fusarium, Acremonium and Geotrichum.

The slaughter house under examination lies in shahaat historical town, El-Gabal El-Akhdar province (Green mountain province) in Libya. Mycological investigations of air in Libyan slaughter houses have not yet been conducted. Therefore the aim of the present study was to investigate air in this slaughter house and throw light on its airborne fungi. The main task was also to look for a standard equation for the rate of sedimentation of mould spores on different surfaces.

MATERIAL AND METHODS

The air of different compartments of shahaat abattoir was investigated mycologically by sedimentation technique recommended by De Boor et al., (1978). Wallhaeuser (1984) and Mansour (1986). Samples were taken from each compartment at different locations. These compartments include slaughtering bleeding dressing, eviceration refrigeration accommodation, evacuation, of intestinal and ruminal contents, and processing (deboning rooms). Examination of lairage, plateform and offices was also peformed. A total of 80 perti-plates (diameter 10 cm) containing mycological agar (Difco) were opened and left for 5 minutes at different locations of the compartments (10 plates in each) during working houre in the morning. Exposure time was 5 minutes to obtain the minimal spreading and at the same time optimal growth. After exposure time the plates were covered tightly with taesa film to prevent further contamination, transported directly to the laboratory, incubated at 22-25°C for 7-10 days according to Samson et al., (1981).

Isolation and identification of moulds depended upon macroscopic and microscopic picture of colonies after one or more purifications (Raper et al., 1965; Ellis, 1971 & 1976; Samson et al., 1976; Domsch et al., 1980; Samson et al. 1981 and Baily and Scott 1985).

RESULTS AND DISCUSSION

It is evident from Table (1) that 360 mould isolates were recovered from all compartments of the examined abattoir. An interesting point of view is that Dematicaeous hyphomycetes (dark moulds) constituted 87.22% of the total isolates under which 7 differ genera were identified namely Cladosporium (56.67%), Alternaria (12.78%), Stemphylium (6.94%), Phoma,

(6.67%), Epicoccum (1.67%), Ulocadium (1.39%) and Phialophora (1.11%). Other airborne fungi were also isolated as Aspergillus (6.67%), Penicillium (2.22%), Fusarium (1.94%). Mucor and Absidia were isolated at low percentages. It is interesting to note from the same table dark moulds constituted over three quarters of the isolates. The achieved results did not agree with those obtained by Hamdy et al., (1990) and Refai et al., (1993). The former authors stated dematiaceous moulds constituted nearly one third of airborne fungi in camel and cattle slaughter halls, while the latter recorded that dematiaceous moulds constituted over a quarter of the isolates (26.41%) of airborne fungi in the modern Egyptian abattoirs . However, in Munich abattoir the results recovered by Mansour, 1986) the dematiaceous hyphomycetes were nearly the same (26.56%) as those obtained by Refai et al., (19933). The results in the present work was not expected . It may be attributed to climatic and geographical variations in Munich, Cairo and Shahaat cities especially the later city locates about 750 meters altitude. It was noticed also from the same table that most of the airborne fungi were isolated from the free compartments in the abattoir such as lairages, dispatcharea as well as slaughter area, to which the living animals arrived with their contaminated coat, dust particles and mud. Dealing with theanimal in slaughter area makes air contaminated with mould spores originated from the aformentioned sources, especially the skin of slaughtered sheep (Mansour, 1986).

Eviceration corners and rooms for evacuation of intestinal and ruminal contents came at the 2nd position, in which airborne fungi were isolated at percentages of 12.78% and 12.22% respectively. Intestinal contents were considered to be the main source of mycological contamination of carcases (Rolle and Kolb, 1954; Klare, 1970; Abdel Rahman, 1981; Mansour et al., 1990). Air may be contaminated by mould spores elaborated from intestinal content particles. This opinion may explain the nearly same number of airborne fungi isolated from eviceration corners and rooms for evacuation of intestinal and ruminal contents. In the same Table (1) a reasonable number of airborne fungi (8.89%) was also recovered from offices of inspector and others dealing with excution of animal slaughtering, dressing and eviceration.. It was obvious that lower numbers of airbornefungi were isolated from refrigeration accomodation and deboning rooms.

Dematiaceous hyphomycetes (dark moulds) are shown in Table (2). A total of 314 (87.22%) isolates were recovered. Cladosporium were the predominating (64.97%), Alternaria (14.65%), Stemphylium (7.96%) and Phoma (7.64%). Epicoccum, Ulocldium and Phialophora were also isolated. Predominance of genus Cladosporium in air was recorded by Morrow et al. (1942), Harris and Chairman, 1950; Gregory, 1954; Kramer et al., 1959; Hudson, 1969; Mansour et al., 1990, which could be attributed to heavy production of spores during the night and spreing of them in the morning (Rich and Waggoner, 1962). However, dematiaceous moulds came on second position of airborne fungi in Munich and modern Egyptian abattoirs (Mansour, 1986; Refai et al., 1993). Intestinal contents act as an important reservoir for black moulds which contaminate floors and air (Mansour et al., 1990).

The results represented in Table (3) show that five spreading were recovered; C. cladosporioids, C. herbarum, C. sphaerospermum, C. macrocarpum, and C. tenuissimum. About a half of the Cladospora were C. cladosporioides, one third C. herbarum and the rest was other Cladospora. Cladosporium cladosporioides and C. herbarum were the most common cladospora isolated from frozen meat in the last 80 years (Talayract, 1901; Berger, 1912, Brooks and Hamsford, 1923; Empay and Scott, 1939; Bate-Smith and Morris,

Table (1): Frequency of airborne fungi at Shahaat abattoir

	Slaughter R*		Evicera-tion R*		Process-ing R*		Refrigera-tor		Offals		Plate form		Lairage		Offices		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Dematiaceous hyphomycetes																		
Cladosporium:																		
C. cladosporioides	14	23.33	12	26.09	2	20.0	-	-	8	18.18	40	40.82	19	31.67	12	37.50	107	29.72
C. herbovirum	12	20.00	4	8.70	6	60.0	2	20.0	8	18.18	14	14.29	11	18.33	6	18.75	63	17.50
C. sphaerospermum	4	6.67	6	13.04	-	-	-	-	-	-	-	-	3	5.0	-	-	13	3.61
C. macrocarpum	2	3.33	2	4.35	-	-	-	-	4	9.09	-	-	-	-	4	12.50	12	3.33
C. tenuissimum	2	3.33	4	8.70	-	-	-	-	-	-	-	-	1	1.67	2	6.25	9	2.50
Total	34	56.67	28	60.87	8	80.0	2	20.0	20	45.45	54	55.10	34	56.67	24	75.0	204	56.67
Alternaria:																		
A. atternata	8	13.33	2	4.35	-	-	-	-	8	18.18	6	6.12	15	25.0	-	-	39	10.83
A. sonchi	-	-	4	8.70	-	-	2	20.0	-	-	-	-	1	1.67	-	-	7	1.94
Total	8	13.33	6	13.04	-	-	2	20.0	8	18.18	6	6.12	16	26.67	-	-	46	12.78
Stemphylium	6	10.0	2	4.35	-	-	-	-	4	9.09	8	8.16	5	8.33	-	-	25	6.94
Phoma	-	-	-	-	-	-	-	-	-	-	24	24.49	-	-	-	-	24	6.67
Epicoccum	-	-	6	13.04	-	-	-	-	-	-	-	-	-	-	-	-	6	1.67
Ulocladium	-	-	-	-	-	-	-	-	4	9.09	-	-	1	1.67	-	-	5	1.39
Phialophora	-	-	4	8.70	-	-	-	-	-	-	-	-	-	-	-	-	4	1.11
Total	48	80.0	46	100	8	80.0	4	40.0	36	81.82	92	93.88	56	93.33	24	75.0	314	87.22
Aspergillus																		
A. flavus	2	3.33	-	-	-	-	-	-	8	18.18	-	-	-	-	-	-	10	2.78
A. fumigatus	-	-	-	-	-	-	-	-	-	-	4	4.08	-	-	2	6.25	6	1.67
A. niger	4	6.67	-	-	2	20.0	-	-	-	-	-	-	-	-	-	-	6	1.67
A. repens	-	-	-	-	-	-	2	20.0	-	-	-	-	-	-	-	-	2	0.56
Total	6	10.0	-	-	2	20.0	2	20.0	8	18.18	4	4.08	-	-	2	6.25	24	6.67
Penicillium																		
P. expansum	2	3.33	-	-	-	-	2	20.0	-	-	-	-	-	-	2	6.25	6	1.67
P. frequentans	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	6.25	2	0.56
Total	2	3.33	-	-	-	-	2	20.0	-	-	-	-	-	-	4	12.50	8	2.22
Fusarium																		
F. oxysporium	-	-	-	-	-	-	2	20.0	-	-	-	-	-	-	2	6.25	4	1.11
F. sporotrioides	-	-	-	-	-	-	-	-	-	-	-	-	3	5.0	-	-	3	0.83
Total	-	-	-	-	-	-	2	20.0	-	-	-	-	3	5.0	2	6.25	7	1.94
Mucor	4	6.67	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4	1.11
Absidia	-	-	-	-	-	-	-	-	-	-	2	2.04	1	1.67	-	-	3	0.83
Total	60	16.66	46	12.78	10	2.78	10	2.78	44	12.22	98	27.22	60	16.67	32	8.89	360	100

R : Room

Table (2): Frequency and distribution of dark mould in air at Shahaat abattoir

	Slaughter R*		Evicera-tion R*		Process-ing R*		Refrigera-tor R*		Offals		Plate form		Lairage		Offices		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Cladosporium																		
C. cladosporioides	14	29.17	12	26.09	2	25.0	-	-	8	22.22	40	43.48	19	33.93	12	50.0	107	34.08
C. herbarum	12	25.0	4	8.70	6	75.0	2	50	8	22.22	14	15.22	11	19.64	6	25.00	63	20.06
C. sphaerospermum	4	8.33	6	13.03	-	-	-	-	-	-	-	-	3	5.35	-	-	13	4.14
C. macrocarpum	2	4.17	2	4.35	-	-	-	-	4	11.11	-	-	-	-	4	16.67	12	3.82
C. tenuissimum	2	4.17	4	8.70	-	-	-	-	-	-	-	-	1	1.79	2	8.33	9	2.87
Total	34	70.83	28	60.87	8	100.0	2	50	20	55.56	54	58.70	34	60.71	24	100	204	64.97
Alternaria																		
A. alternata	8	16.67	2	4.35	-	-	-	-	8	22.22	6	6.52	15	26.79	-	-	39	12.42
A. sonchi	-	-	4	8.70	-	-	2	50	-	-	-	-	1	1.79	-	-	7	2.23
Total	8	16.67	6	13.04	-	-	2	50	8	22.22	6	6.52	16	28.57	-	-	46	14.65
Stemphylium	6	12.50	2	4.35	-	-	-	-	4	11.11	8	8.70	5	8.93	-	-	25	7.96
Phoma	-	-	-	-	-	-	-	-	-	-	24	26.09	-	-	-	-	24	7.64
Epicoccum	-	-	6	13.04	-	-	-	-	-	-	-	-	-	-	-	-	6	1.91
Ulocladium	-	-	-	-	-	-	-	-	4	11.11	-	-	1	1.79	-	-	5	1.59
Phialophora	-	-	4	8.70	-	-	-	-	-	-	-	-	-	-	-	-	4	1.27
Total	48	15.29	46	14.65	8	2.55	4	1.27	36	11.46	92	29.30	56	17.83	24	7.64	314	

R : Room

Table (3): Frequency of isolated cladosporium species in air at Shahaat abattoir.

Cladosporium species	Sl. halls		evicerat. rooms		Process, rooms		Refri. rooms		Offals rooms		Plate form		Lairage		Offices		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
C. cladosporioides	14	41.18	12	42.86	2	25.00	-	-	8	40.00	40	74.07	19	55.88	12	50.00	107	52.45
C. herbarum	12	35.29	4	14.29	6	75.00	2	100	8	40.00	14	25.93	11	32.35	6	25.00	63	30.88
C. sphaerospermum	4	11.76	6	21.43	-	-	-	-	-	-	-	-	3	8.82	-	-	13	6.37
C. macrocarpum	2	5.88	2	7.14	-	-	-	-	4	20.00	-	-	-	-	4	16.67	12	5.88
C. tenuissimum	2	5.88	4	14.29	-	-	-	-	-	-	-	-	1	2.94	2	8.33	9	4.41
Total	34	16.67	28	13.73	8	3.92	2	0.98	20	9.80	54	26.47	34	16.67	24	11.76	204	100

Table (4): Comparison between the revealed airborne fungi at different abattoirs

Isolated moulds	Germany, Munich. Mansour, 1986		Egypt				Libya/ Shahaat abattoir	
			Old Cairo abattoir Hamdy et.. al 1990		Modern abattoir refai et. al 1993			
	No.	%	No.	%	No.	%	No.	%
Dematiaceous moulds	81	26.56	257	31.89	564	26.40	314	87.22
Cladosporium	36	11.80	-	-	317	14.84	204	56.67
Alternaria	43	14.10	-	-	125	5.85	46	12.78
Stemphylium	-	-	-	-	12	0.56	25	6.94
Phoma	-	-	-	-	-	-	24	6.67
Epicoccum	-	-	-	-	9	0.42	6	1.67
Ulocladium	1	0.33	-	-	-	-	5	1.39
Phialophora	-	-	-	-	-	-	4	1.11
Curvularia	-	-	-	-	25	1.17	-	-
Helminthosporium	-	-	-	-	56	2.62	-	-
Exophialia	1	0.33	-	-	-	-	-	-
Nigrospora	-	-	-	-	11	0.51	-	-
Stachybotrys atra	-	-	-	-	9	0.42	-	-
Aspergillus	90	29.51	339	42.06	703	32.91	24	6.67
Penicillium	34	11.15	134	16.63	430	20.13	8	2.22
Paccilomyces	-	-	1	0.12	5	0.23	-	-
Scopulariopsis	12.	3.93	3	0.37	4	0.19	-	-
Absidia	11	3.61	8	0.99	-	-	3	0.83
Mucor	7	2.30	6	0.74	149	6.98	4	1.11
Rhizopus	4	1.31	19	2.36	50	2.34	-	-
Fusarium	4	1.31	5	0.62	114	5.34	7	1.94
Acremonium	2	0.66	8	0.99	13	0.61	-	-
Geotrichum	2	0.66	2	0.25	68	3.18	-	-
Trichoderma	1	0.33	24	2.98	14	0.66	-	-
Thamnidium	1	0.33	-	-	-	-	-	-
Trichophyton	1	0.33	-	-	-	-	-	-
Candida	-	-	-	-	22	1.03	-	-
Others	55	18.03	-	-	-	-	-	-
Total	305		806		2136		360	
Sedimentation rate	0.01		0.02		0.01		0.01	

1952; Frazier and Westhoff, 1979; Lowry and Ashton; 1982; Mansour et al., 1991). The same two species of Cladosporium were incriminated in the formation of black spots on chilled and frozen meat, where they are able to grow and produce this affection till-12°C (Gill et al., 1981; Michener and Elliot 1964; Noskowa, 1975; Mansour, 1986). There is a correlation between the rate of people infection who suffered from pulmonary affections and airborne fungi (Refai et al., 1993). Other moulds as Aspergillus, Penicillium and Fusarium were isolated at low percentages (Table 1). The recovered results showed high level of mould contamination in air. It could be attributed to poor hygienic measures and open environment in abattoir under investigations.

The sedimentation rate of colony forming unit (CFU) of airborne fungi per one cm^2 per one minute could be calculated according to this new equation:

$$R = \frac{N}{nE_t \ \Pi r^2}$$

Where

R: Sedimentation rate of airborne fungal spores. per cm^2 per minute.
N: Total number of isolated moulds.
n: Number of plates used.
Et: Exposure time in minutes.
II: Constant and equal to 3.14.
r: Radius of the plate.

According to this new on the sedimentation rate of airborne fungi one cm^2/minute is 0.01 in Shahaat abattoir. After calculation it was 0.01, 0.02 and o.01 in Munich, old Cairo and modern Egyptian abattoirs (Mansour; 1986; Hamdy et al., 1990 and Refai et al., 1993 respectively). Sedimentation rate of airborne fungi in slaughter houses could be obtained according to this equation. Furthermore, limits for the degree of air contamination in abttoirs, food processing plant and food serving establishments ..... etc., may be tabulated and qualified in the future. The colony forming unit CFU/m^3 of air obtained by different types of air samples (Mulhausen et al., 1987) may be also tabulated, evaluated and compared with the results recovered by this new equation.

To minimize the airborne fungi in the abattoir, strict hygienic measures should be applied as recommended by FAO such as coordination of movement in the abattoir, enterence of slaughtered animals and exit of carcases. Animals must be rested at least 24 hours in lairages before slaughtering. The most important is washing of animals with dushes and periodical cleansing of slaughter halls, removal of dirts and dusty materials. Induction of educational programmes for workers dealing with animals and meat to ensure safety of meat and environment.

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