قســـــــم : الصحـة ومراقبـة الأغذية - كلية الطب البيطرى - جامعة القاهرة
رئيس القســم : أ.د / محمـد عبدالرحمن عشـــوب

دراسات ميكروبيولوجية عن البيض الفاســـــد

عبد الوهاب مرسى ، عبـده العشـماوى ، اخلاص مرســـــى

جمعت عينات من بيض الدجاج المتبقى أو المخزون بمحلات بيع البيض فى القاهرة وضواحيها حيث أجريت عليهـــا الفحـوص الظـاهرية والميكروبيولوجيـــة .

دلت الفحوص الميكروبيولوجية عن وجود ٤٢ نوع من الفطريات والخمائر وكذلك ١٢١ نوع تتبع أنواع مختلفة من البكتريا .

أثبت التصنيف السيرولوجى لميكروبات السالمونيلا المعزولة عن وجود ٧ أنواع بنسب تتراوح بين ٤ر٣٪ الى ٨٢ر٣٣٪ وكانت العترات المعزولة حسب الترتيب التنازلى الآتى : سالمونيلا بللورم ، سالمونيلا توسون ، سالمونيلا نيوبورت ، سالمونيلا تيفـى ميوريم ، سالمونيلا كيفو ، سالمونيلا جورجيا وسالمونيلا أمهيلالى .

تم عزل ٢٦ ميكروبا من ايشيريشيا كولاى ، أمكن تصنيف ٢١ منها سيرولوجيا ، كذلك تم عزل ميكروبات البروتيس ، سيد ومونـاس ابروجينوزا ، كلبسيلا ، انتيروباكتركلوكا والكاليجينس فيكالس بنسبة ٠٩ر١٩٪ ، ١٨ر١٨٪ ، ٩١ر١٠٪ ، ٠٩ر٩٪ و ٢٧ر٧٪ على التوالى .

أمكن عزل الفطريات وتصنيفها وجد أنها تتبع أنواع اسبرجلس ، بنيسليم ، ريزوبس بنسب تتراوح بين ٣٧ر٩٪ الى ٨٧ر٢١٪ وكذلك تم عزل الخميرة بنسبة ٠٩ر٩٪ وتصنيفها وجدت أنها تتبع نوع كاندايدا البيكانز .

تناولت الدراسة مناقشة أهمية الميكروبات المعزولة وعلاقتها بالصحة العامة وكذلك اقتراح السبل الكفيلة بالمحافظة على البيض من التلوث بالميكروبات المختلفة ومنع تكاثرها أثناء التخزين حفاظا على البيض من الفساد وحماية للمستهلك من الاصابة بالامراض.

Dept. of Hygiene and Food Control,
Faculty of Veterinary Medicine, Cairo University,
Head of Dept. Prof. Dr. M.A. Ashoub.

MICROBIOLOGICAL STUDIES ON DETERIORATED HEN EGGS

(With 5 Tables)

By

A.W. MOURSY; A.M. AL-ASHMAWY and E.A. MOURSY

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SUMMARY

Unsold aged hen eggs, collected from different egg shops in Cairo and its Suburbs were examined microbiologically.

Group analysis of the results showed that 42 contaminants belonging to different types of moulds and yeasts as well as 121 bacterial isolates could be identified.

The serological typing of isolated Salmonellae revealed the presence of 7 serotypes, with an incidence percentage ranging from 4.17% to 33.12%. Isolated strains in a descending manner included S. pullorum, S. thompson, S. New port, S. typhimurium, S. Kivo, S. goergia and S. Umhelali.

Out of the 26 isolated Escherichia Coli, only 21 isolates could be typed serologically (eleven serotypes).

Proteus species, Pseudomonas aerogenosa, Klebsiella, Enterobacter coloaca and Alcaligenes feacalis could be isolated from contaminated eggs with an incidence percentage of 19.09%, 18.18%, 10.91%, 9.09% and 7.27% respectively.

The frequency destribution of isolated moulds revealed that Aspergillus fumigatus was the most prevalent (21.87%) while Penicillium oxalicum was the least (9.37%) and Aspergillus flavous, Aspergillus niger, Penicillium cycolabium, Penicillium chrysogenum and Rhizopous lie inbetween. Isolated yeasts proved to be Candida albicans (9.09%).

The economic and public health importance of isolated microorganisms has been discussed.

Suggested measures for improving the quality of produced hen eggs are given.

INTRODUCTION

The value of eggs in human nutrition is high not only for body maintenance, but also for growth, lactation and reproduction as it presents a good source of high quality animal protein and other food elements.

Although it is generally agreed that microbial flora of hen eggs at the time of laying is very few, if any, yet contamination of eggs after laying with a variety of organisms from different sources exists.

Spoilage of hen eggs due to the activity of different types of contaminants that find their way to the egg contents as a result of faulty production, handling or storage causes economic losses. Moreover contaminated eggs may, at times, constitute a public health hazard.

Studies on existing microorganisms in deteriorated hen eggs in our country is very scanty and limited. Therefore, this work has been planned to investigate the different types of microorganisms prevailing in deteriorated hen eggs.

MATERIAL AND METHODS

Unsold aged hen eggs collected from different eggs shops in Cairo and its Suburbs were transferred to the laboratory where they were subjected to physical examinations to detect abnormal eggs, which are to be examined microbiologically as follows:

Preparation of eggs: Eggs were cleaned and sterilized externally before a sufficient area fo the shell was removed around the air sac.

Experimental procedures:

Moulds and yeasts:

Loopfuls from the inner shell membrane as well as from the mixed egg contents were directly streaked on Sabouraud maltose agar medium, containing 0.5 mg of chloramphinicol per ml. Inoculated plates were incubated for 5

days at 25°C before being examined. Suspected colonies were isolated on Sabouraud's medium for further identification according to AJELLO and GEORE(1964) for isolated moulds and LODDER & KREGER-VAN RIJ(1952) for isolated yeasts

Salmonellae:

Heavy inoculum from the mixed egg contents was transferred to Selenite F broth (Difco) before being incubated at 37 C for 18 hours, after which loopfuls were streaked on three specific selected solid media (MacConkey, Brillient green and S.S. agar, "Difco"). Inoculated plates were incubated at 37°C for 24 hours before being examined. After incubation suspected Salmonella growth, according to their characteristics, were examined microscopically before being isolated in pure culture on agar slope for further identification according to EDWARDS and EWING (1972). The seriological typing of isolates was done in the Institute of Animal Health, Dokki, Giza, ARE.

Proteus spp.:

Swarming growth on the plating media was further tested for identification according to EDWARDS & EWING (1972).

Pseudomonas spp.:

Suspected Pseudomonas growth on nutrient agar plates was identified biochemically according to EDWARDS & EWING (1972).

Alcaligenes faecalis:

Non-lactose fermenting isolates on MacConkey's agar plates showing positive motility test and growth on Simmon's Citrate agar were further subjected for biochemical identification according to EDWARDS & EWING (1972).

Coliform organisms:

Pure culture from suspected growth on specific media were identified biochemically and serologically according to EDWARDS & EWING (1972).

RESULTS

The results of microbiological examination were recorded in Tables (1 - 5).

Group analysis of isolates shows that 42 contaminants belonged to different types of moulds and yeast, while 121 isolates belonged to different types of bacteria.

DISCUSSION

A- Bacterial Contaminants:

Salmonellae:It's evident from the results given in table (1) that 24 Salmonella organisms representing an incidence percentage of 21.82, could be isolated from contaminated eggs. The serological typing reveals that isolated strains included S. pullorum, S. thompson, S. newport, S. typhimurium, S. kivo, S. georgia and S. umbelali in a descending manner (Table 2). These findings substantiate what have been reported by EL-AGROUDY and AWAD (1966), AHMED (1969) and MOURSY & AHMED (1971).

Salmonellosis is widely spread among birds, specially among water birds. In adult birds, latent form is metwith, which is dangerous from epidemiological point of view, as they remain for a long time laying infected eggs and spreading infection among the flock. Different types of Salmonellae from soiled egg shell with infected material can penetrate into the egg and find their way directly to the yolk where they grow. The rate of penetration depends on the holding temperature (SIMONS et al., 1970; MOURSY & AHMED, 1971 and SOUTER & PETERSON, 1974). Moreover, Salmonellae could be isolated from frozen or dried eggs by different investigators (FRAZIER & WESTHOFF, 1978).

From the public health point of view, Salmonellae infection as well as food poisoning outbreaks have been attributed to consumption of hen eggs, or egg products (TAYLOR, 1969 and McCOY, 1975).

Escherichia Coli: Inspection of table (1) reveals that Escherichia coli could be isolated 26 times (23.64%) from examined eggs. The frequency distribution of isolated strains, based on serological typing,is recorded in table (3). These findings are in agreement with those reported by BOARD & BOARD (1968), AHMED (1969) and AHMED et al., (1974).

Coliform organisms are considered among contaminants of rotten eggs (SEVIOUR et al., 1972) and certain strains of Escherichia coli produce fishy flavours (FRAZIER & WESTHOFF, 1978). Members of entero-pathogenic serotypes of Escherichia coli have been implicated in human cases of gastroenteritis, epidimic diarrhaea in infants and sporadic summer diarrhaea in children (MACKIE & McCARTNEY, 1962).

Proteus species: could be isolated from 21 eggs (19.09%). Identification of isolated Proteus organisms given in table (4) points out that Proteus vulgaris was the most prevalent species (57.1%), while Proteus rettgeri was the least (14.2%). Species of proteus could be isolated from rotten eggs by different authers (BOARD, 1965; BOARD & BOARD, 1968 and AHMED et al., 1974).

Proteus organisms cause economic losses through deterioration of eggs even when kept at low temperature (FRAIZER & WESTHOFF, 1978). Moreover, some of isolated species constitute a public health hazard (MACKIE & McCARTNEY, 1962).

Pseudomonas aerogenosa: could be isolated from 20 eggs (Table 1). This finding substantiates what has been reported by BOARD (1965); BOARD & BOARD (1968); AHMED (1969) and AHMED et al. (1974). Pseudomonas species are among the Gram-negative bacteria which induce changes in the edible constituents of egg, correlated with the biochemical properties of the organism. Such organisms have been implicated in different kinds of bacterial egg rot resulting in objectionable changes rendering the eggs unfit for human consumption (FRAZIER & WESTHOFF, 1978).

Alcaligenes faecalis: was isolated from 8 eggs (7.23%). Nearly similar finding was reported by BOARD (1965). On the other hand higher percentage was recorded by AHMED (1969). Isolated organism besides being responsible for deterioration of eggs, it has a public health importance as it has been isolated from cases of enteritis (MACKIE & McCARTNEY, 1962).

Klebsiella and Enterobacter Coloaca: could be isolated from examined eggs with an incidence percentage of 10.9% and 9.09% respectively (Table 1). These findings substantiate what has been reported by AHMED (1969). Enterobacter coloaca was isolated from rotten eggs by BOARD (1965) and BOARD & BOARD (1968). Enterobacter coloaca has been considered as a secondry invador producing spoilage eggs (FRAZIER & WESTHOFF, 1978).

B. Moulds and Yeasts:

The percentage distribution of isolated micro-organisms from contaminated eggs given in table (1) shows that moulds were more prevailing (29.09%) than yeasts (9.09%). The frequency distribution of isolated fungi reported in table (5) reveals that seven different species of moulds could be identified. Aspergillus fumigatus was the most prevalent (21.88%); while Penicillium oxolicum was the least (9.38%). Isolated yeasts proved to be Candida albicans.

The spoilage of eggs by fungi was reported by FRAZIER & WESTHOFF (1978). On the other hand some of isolates were recovered from rotten eggs by AHMED et al., (1974).

It is worth to mention that rotten eggs metwith in this work contained mixed infection of mostly Gram-negative bacteria including members of the enteric group. A finding indicative that such organisms have gained access to egg contents from soiled shells with body wastes, or sometimes directly from infected birds or carriers.

The results achieved allow to conclude that the incidence of contaminants in unsold aged market eggs is comparatively high resulting in economic losses, besides constituting a public health hazard.

Realizing that the quantity of produced eggs in our country is insufficient to meet the consumers demand specially in summer time when production drops and the hygienic methods of handling, storage and distributiôn has not yet been established. Therefore, rigid attention to every detail of plant hygiene and correct treatment and storage are essentials for satisfactory production.

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TABLE (1)

Frequency distribution of isolated microorganisms from examined eggs (110 eggs)

Isolated organisms	No of isolates	%
Salmonellae	24	21.82
Proteus species	21	19.09
Pseudomonas aeroginosa	20	18.18
Alcaligenes feacalis	8	7.27
Escherichia Coli	26	23.64
Klebsiella species	12	10.91
Enterobacter Coloaca	10	9.09
Moulds	32	29.09
Yeasts	10	9.09
Total	163	

TABLE (2)

Frequency distribution of isolated Salmonellae based on their serological typing

Isolated Salmonellae	Frequency	
	No	%
Salmonella newport	3	12.50
Salmonella Umhelali	1	4.17
Salmonella Kivo	2	8.33
Salmonella pullorum	8	33.32
Salmonella thompson	5	20.85
Salmonella georgia	2	8.33
Salmonella typhimurium	3	12.50
Total	24	100.00

TABLE (3)

Frequency distribution of isolated Escherichia Coli based on their serological typing

Isolated organisms		Frequency	
		No	%
Escherichia Coli	0101	2	7.69
Escherichia Coli	099	2	7.69
Escherichia Coli	0119	2	7.69
Escherichia Coli	0123	1	3.85
Escherichia Coli	011	3	11.54
Escherichia Coli	08	2	7.69
Escherichia Coli	022	2	7.69
Escherichia Coli	053	1	3.85
Escherichia Coli	017	1	3.85
Escherichia Coli	0111	2	7.69
Escherichia Coli	048	3	11.54
Untypted		5	19.23
Total		26	100.00

TABLE (4)

Frequency distribution of isolated species of Proteus organisms

Isolated organisms	Frequency	
	No	%
Proteus vulgaris	12	57.14
Proteus rettgeri	3	14.29
Proteus mirabilus	6	28.57
Total	21	100.00

TABLE (5)

Frequency distribution of isolated Moulds and Yeasts

Isolated organisms	Frequency	
	No	%
Aspergillus flavous	5	15.63
Aspergillus niger	5	15.63
Aspergillus fumigatus	7	21.87
Penicillium Cycolobium	4	12.50
Penicillium Oxalicum	3	9.37
Penicillium Chrysogenum	4	12.50
Rhizopus	4	12.50
Total	32	100.00
Candida albicans	10	100.00