قسم: المراقبة الصحية على الأغذية كلية: الطب البيطري \_ جامعة أسيوط رئيس القسم: أحد • توفيق البسيوني •

مدى تواجد الميكروبات المعوية في بعض أنواع الأغذية

أحمد عبدالحميد ، حسني عبداللطيف\*، مصطفى خليل

أجريت هذه الدراسة على ١٢٠ عينة من اللبن الخام ، الجبن الدمياطيوالقريش والهامبورجر والسجق جمعت عشوائيا من أسواق مدينة أسيوط والسوبر ماركت ومحللات المعاللة وذلك لمعرفة مدى تواجد الميكروبات المعوية ٠

وقد تبين من الدراسة أن ۷۰، ۳۵، ۸۰، ۹۰، ۳۵٪ من العينات المفحوصة علي وقد تبين من الدراسة أن ۷۰، ۳۵، ۳۵٪ من العوية Enterobacteriaceae بينما وجد أن ميكروب الـ Proteus , E. coli تم عزلها على التوالي من ۷۰، ۳۰٪ من اللبن الخام، ميكروب الـ بالجبن الدمياطي، ۸۰ و ۱۲٪ من الجبن القريش، ۱۰٫۸۵٪ من الجبن المونيلا من ۱۰، ۵٪ الهامبورجر ، ۵۰ و ۱۰، ۵٪ من السجق وقد تم عزل ميكروب السالمونيلا من ۱۰، ۵٪ من عينات الجبن الدمياطي القاريش والسجق بالترتيب وقد تم عزل مينات الجبن الدمياطي القاريش والسجق بالترتيب

كذلك تم تصنيف مجموعة ال Enterobacteriaceae والتي تم عزلها منالعينات وهي كالتالي:

Enterobacer hafniae & Liquefaclent; Citrobacter diversus & Freundii; Alkaligenes faecalis; Klebsiella spp., Providencia spp.; Edardsiella tarda; Proteus (Mirabilis, rettergi & vulgaris); Acinetobacter spp., Serratia Liquefaciens & Marcescens, Arizona hinshawii and Pseudomonas aerogenosa.

ولقد تم مناقشة خطورة هذه الميكروبات على الصحة العامة وكذلك الشروط التـــي يجب توافرها لمنع تلوث الأغذية بهذه الميكروبات٠

<sup>\*:</sup> قسم الصحة \_ كلية الطب البيطري \_ جامعة أسيوط.

Dept. of Food Hygiene, Fac. of Vet. Med., Assiut University, Head of Dept. Prof. Dr. T.A. El-Bassiony.

# INCIDENCE OF ENTEROBACTERIACEAE IN SOME SELECTED FOOD STUFFS

(With 3 Tables)

AHMED A-H. AHMED; H.A. ABDEL-RAHMAN\* and M.K. MOUSTAFA (Received at 3/3/1988)

# SUMMARY

One hundred and twenty random samples of some selected food items, including, raw milk, Damietta and Kareish cheese, hamburger and fresh sausage, collected from different localities in Assiut City, were sereened for fEnterobacteriaceae. The obtained results revealed that 70, 35, 80, 90 and 35% of the examined samples, respectively, were contaminated by Enterobacteriaceae. The different isolates recovered from the examined food samples were identified as E.coli; Enterobacter hafniae and liquefacient; Citrobacter diversus and freundii; Alkaligenes faecalis; Klebsiella ssp.; Providencia spp.; Endwardsiella tarda; Proteus mirabilis, rettergi and Vulgaris; Acinetobactr spp.; Serratia liquefaciens and marcessens; Salmonella spp.; Arizona hinshawii and Pseudomonas aerogenosa. The public healths and suggestive measures were discussed.

## INTRODUCTION

The safety of food depends on its freedom from bacteria known to cause food poisoning and also from mass bacterial contamination resulting from careless storage. Coliform bacteria have received more attention than most other groups of bacteria occurring in foods; they have been used as indicators of unsatisfactory manufacturing practices. More recently determination of any or all members of the family Enterobacteriaceae as a microbiological guide line and indicator of food quality and sanitation has attracted the attention of several food scientists.

Many species of family Enterobacteriaceae may constitute, if contaminate the food, a public health hazard. Many cases of food poisoning outbreaks were traced to the members of family Enterobacteriaceae (RIDGE and THOMAS, 1955; BREED et al., 1957; THOMASON et al., 1961; MAKIE and MacCARTNEY; 1962; FRAZIER, 1967; IRODANOV et al., 1970; EDWARD and EWING, 1972; TULLOCH et al., 1973; FINEGOLD and MARTIN, 1974; FANTASIA et al., 1975 and HOBBS and GILBERT, 1978). Different food items including milk, cheeses, hamburger and sausage were screened for Enterobacteriaceae by several investigators: HALL et al. (1967); MOUSTAFA et al. (1975); EL-BASSIONY (1977); SHELAIH (1979); SAUDI (1980); HEFNAWY (1980); EL-KHATEIB (1982) and GADEL-RAB (1983).

Therefore, this work was planned to screen some selected food stuffs available at retail outlets in Assiut City for the members of Enterobacteriaceae to assess their quality and sanitation.

<sup>\* :</sup> Dept. of Food Hygiene, Faculty of Vet. Med., Suez Canal Univ.

# AHMED A-H. AHMED, et al.

# MATERIAL and METHODS

# Collection of samples:

A total of 120 random samples of some selected food items, including, 20 each of raw milk and Domietta cheese, 40 kareish cheese, 20 each of frozen hamburger and fresh sausage were collected from Assiut City markets, different groceries and supermarkets. Raw milk samples were obtained from different dairy farms in Assiut City. The samples were dispatched to the laboratory with a minimum of delay.

# Preparation of the samples:

Milk and cheese samples were handled and prepared for examination according to standard methods for the examination of dairy products (A.P.H.A., 1978). Hamburger and fresh sausage samples were aseptically freed from their casing and thoroughly ground, mixed and homogenized using sterile blenders. All samples after preparation were subjected to the following examination:

- 1- Detection and identification of salmonella and shigella organisms according to the methods recommended by SPECK (1976).
  - 2- Isolation and identification of E.coli according to SPECK (1976).
  - 3- Isolation and identification of other members of Enterobacteriaceae.

25 g of the afore mentioned prepared food samples were aseptically inoculated into 225 ml selenite cystine broth. Inoculated broth flasks were incubated at 37°C for 24 h. Loopfuls of 24 h enrichment media were streaked on Brillient green and MacConkey's agar plates in a way to obtain separate colonies, and incubated at 37°C for 24 h. Different colonies of both lactose and non lactose formenting bacteria were picked up onto agar slants for further purification and identification according to EDWARDS and EWING (1972) and FINEGELD and MARTIN (1974).

Serological identification of strains presumed to be salmonella or shigella was carried out in the Dept. of Bacteriology, Faculty of Medicine, Assiut University.

## RESULTS

The obtained results were recorded in Tables 1, 2 and 3.

#### DISCUSSION

Tables 1 & 2 showed that 70, 35, 80, 90 and 85% of the examined raw milk. Damietta and Kareish cheese, hamburger and sausage samples were contaminated by Enterobacteriaceae, respectively. E.coli and proteus spp. could be detected in 70, 30% of raw milk; 20 & 10% of Damietta cheese; 80 & 12.5% of kareish cheese; 85 & 10% of hamburger and 50 & 12.5% of sausage samples, respectively. Higher incidence of Enterobacteriaceae and E.coli, as well as, lower incidence of Proteus spp. were recorded in raw milk, Damietta and kareish cheese samples examined by GADEL-Rab (1983). Also, a lower incidence of E.coli was detected in Damietta and kareish cheese samples examined by EL-BASSIONY (1975) and AHMED et al. (1987), respectively. Salmonella spp. did not recover from the examined raw milk and hamburger samples, while the organism could be isoalted from 5, 10 and 5% of Damietta and kareish cheese and sausage samples, respectively. Such results disagreed with those obtained by ABDEL-RAHMAN (1972); EL-KHATEIB (1982) and GADEL-RAB (1983), who failed to isolate Salmonella spp. from

#### ENTESOBACTERIALEAE IN FOOD

Table (1): Incidence of Enterobacteriaceae in some selected food samples.

Food samples	No. of	Pos	Positive samples					
	samples	No	000					
Raw milk	20	14	70					
Damietta cheese	20	. 7	35					
Kareish cheese	40	32	80					
Hamburger	20	18	90					
Sausage	20	. 17	85					
Total	120	88	73.3					

Table (2): Incidence of E.coli, Salmoneela and proteus in the examined food samples.

Food samples		Poitive samples									
		E. co			coli	coli Salmonella			Proteus		
		No.		No.	0/	401	No.	0/0	er mark	No.	00
Raw milk	7350 F 3 150	20	el Yr.	14	70		-	202 F		6	30
Damietta cheese		20		4	20		1	5		2	10
Kareish cheese		40		32	80		2	10		5	12.5
Hamburger	The same	20		17	85		-	-	- E	2	10
Sausage	The Contract of the Contract o	20	district the second	10	50		1	5	7.7	5	12.5
Total		120		77	64.2	THE PARTY OF	4	3.3	one is to	20	16.7

the examined samples of cheeses and sausage. On the otherhand, lower incidence of E.coli and Proteus spp. were detected in hamburger and sausage samples examined by Hefnawy (198)) and EL-KHATEIB (1982), respectively.

Different isolates of family Elknterobacteriaceae could be isolated from the examined food samples (Table 3). Most of these isolates were previously detected in raw milk, cheeses, hamburger and sausage samples examined by GHONEIM (1963); HALL et al. (1967); MOUSTAFA et al. (1975); KUMAR et al. (1978); SHELAIH (1979); SAUDI (1980); HEFNAWY (1980); EL-KHATEIB (1982) and GADEL-RAB (1983). Shigella spp. could not be isolated from the examined food samples. The same finding was obtained by SKOUNTZONS et al. (1973); HEFNAWY (1980); EL-KHATEIB (1982) and GADEL-RAB (1983). Some of the isolated strains, including E.coli, Citrobacter, Klebsiella, Providencia, Proteus and Salmonella have been involved in many cases of food poisoning outbreaks (SMITH and CONANT, 1960; FRAZIER, 1967; IORDANOV et al., 1970; EDWARDS and EWING, 1972; FINEGOLD and MARTIN, 1974 and FANTASIA, 1975).

In conclusion, the results of such investigation proved that the examined food samples were considered seriously contaminated by Enterobacteriaceae organisms, and thus reflex the unsatisfactory measures of food production and handling. Therefore, more care and more governmental supervision should be directed towards the quality and sanitation of foods available at retail outlets. Strict hygienic measures should be imposed during food production and handling to avoid additional outbreaks of food-borne illness caused by such organisms.

AHMED A-H. AHMED, et al.

Isolates	Raw milk		Damietta cheese		Kareish cheese		Hamburger		Susage	
	No.	%	No.	%	No.	%	No	%	No.	%
3. coli	14	41.20	4	26.67	32	56	17	45	10	31.3
Entarobacter :								. **		
hafniae	-	-	-	-	2	3.5	1	2.6	-	
liquefacient	7	20.58	3	20	-		4	10.5	2	6.3
Citrobacter										
diversus	2	5.88	-	- 12	1	1.8	1	2.6		
freundii	. 1	2.94	-	-	3	5.3	1	2.6	1	3.
Alkaligenes faecalis	-	-	3	20	-	-	1	2.6		
Clebsiella sp.	-	-	-	-	-	-	2	5.3	1	3.
Providencia sp.	-	-	1	6.67	1	1.8	1	2.6	2	6.
Edwardsiella Tarda	2	5.88	1	6.67	-	-	1	2.6	1	3.
Proteus										
micrabilis		-	-	-	-	-	-	-	3	9.
rettergi	3	8.82	1	6.67	2	3.5	1	2.6	1	3.
vulgaris	3	8.82	1	6.67	3	5.3	1	2.6	1	3.
Acinetobacter sp.	14 1 2	-			-	-	-	-	1	3.
Serratia										
liquefaciens	1	2.94	-	and the	2	3.5	4	10.5	1	3.
marcescens	1	2.94	-	-	4	7	1	2.6	3	9.
Arizona hinshawii	-	-	-		4	7	2	5.3	4	12.
Salmonella sp.	-	-	1	6.67	2	3.5	-	-	1	3.
Ps. serogenosa	-	-	•	-	1	1.8	a 1, -	-	-	_
Total	34	100	15	100	57	100	38	100	32	10

# REFERENCES

Abdel-Rahman, H.A. (1972): Studies on Egyptian white cheese. M.V.Sc. Tkhesis, Fac. of Vet. Med. Cairo Univ.

Ahmed, A., A-H.; Moustafa, M.K. ad Abdel-Hakiem, E.A. (1987): Sanitary condition of kareish cheese manufactured in Assiut City. Assiut Vet. Med. J. 19 (37): 76-80.

cheese manufactured in Assiut City. Assiut Vet. Med. J. 19 (37): 76-80.

A.P.H.A. (1978): Standard Methods for the examination of dairy products E.H. Marth (ed),

American Public Health Association 14th. Washington D.C.

Assiut Vet.Med.J. Vol. 20, No. 40, 1988.

#### ENTESOBACTERIALEAE IN FOOD

- Breed, S.R.; Smith, N.R. and Murray, E.G.D. (1957): Bergey's Manual of determinative Bacteriology, 7th Ed., Tindal and Cox Ltd. London.
- Edwards, P.R. and Ewing, W.H. (1972): "Identification of Enterobacteriaceae", 3rd Ed. Burgess Bublishing Company, Minneapolis, Minnesota.
- El-Bassiony, T.A. (1975): Studies on karish cheese in Upper Egypt M.V.Sc. Thesis Fac. Vet. Med. Assiut University.
- EI-Bassiony, T.A. (1977): Incidence of pathogenic microorganisms and viability of some foodpoisoning bacteria in kareish cheese durig storage. Ph.D. Thesis, Fact. Vet. Med. Assiut University.
- El-Khateib, T.S. (1982): Sanitary condition of sausage in Assiut. M.V.Sc. Thesis, Fac. of Vet. Med. Assiut University.
- Fantasia, L.D.; Mestrandrea, L., Schrade, J.P. Yager, J. (1975): Detection and growth of enteropathogenic E.coli in soft ripened cheese Appl. Microbial 29: 179-185.
- Finegold, S.H. and Martin, W.J. (1982): Baily and Scott Diagnostic microbiology 6th ed. C.V. Mosby Co. St. Louis, Torento, London.
- Frazier, W.C. (1967): "Food Microbiology" 2nd Ed., New York, Mc Graw Hill. Inc.
- Gadel-Rab, H.M. (1983): Enterobacteriaceae in milk and milk prducts. M.V.Sc. Thesis. Fac. Vet. Med. Assiut Univ.
- Ghoneim, N. (1963): Studies on microflora of Damietta cheese. M.V.Sc. Thesis, Cairo Univ. Hall, H.G.; Brown, D.E. and Ewis, K.H. (1967): Examination of market food for coliforms. Appl. Microbiol., 15, 1062.
- Hefnawy, Y. (1980): Studies on sanitary improvement of processed meat (Hamburger) manufactured in Egypt. Ph.D. Thesis. Assiut Univ.
- Hobbs, B.C. and Gilbert, R.J. (eds) (1978): Food poisoning and Food hygiene 4th Ed. English language Book society and Edward Arnold (puplisher) Ltd.
- lordqnov, I.; Slavkov, I. and Bozhilov, B. (1970): Occurrence of (Salmonella in the mammary gland of) ewe. 1st National conference of Salmonellae and salmonellosis in Bulgaria Lgariya 115. Dairy Sci. Abst. 34, 6.
- Kumar, S.; Sinha, B.K. and Sahal, B.N. (1978): Bacterial quality of the raw buffalo milk marketed in and around Patna and its public health importance. Indian J. Dairy Sci., 31, 2: 156.
- Mackie, K.J. and Maccartney, J.E. (1962): Hand book of practical bacteriology 10th Ed. & S. Livingstone Ltd., London.
- Moustafa, T.H.; Kamel, Y.Y.; Ahmed, A.A. and Ismail, A.A. (1975): Studies on the sanitary condition of market milk in Assiut Province, II- The incidence of potentially pathogenic microorgansims. Assiut Vet. 2 (314)181.
- Ridge, L.E. and thomas, M.E.M. (1955): Infection with Providencia type of paracolon bacillus, J. Path. Bact., 69, 335.
- Saudi, A.M.M. (1980): Microbiological studies on food-poisoning microorganisms in some market dairy products. Ph.D. Thesis Fac. Vet. Med. Cairo Univ.
- Shelaih, M.A. (1979): Microbiological studies on Egyptian soft cheese, Ph.D. Thesis, Fac. Vet. Med. Cairo Univ.
- Skountzos, K.; Papadias, A. and Yiotis, A. (1973): The hygienic condition of Greek cheese, Deltiontes Ellenikes Kteniatrikes Etaireias 24, 3: 143. Dairy Sci. Abst., 35, 11.
- Smith, E.D. and Conant, N.F. (1960): "Zinssor microbiology" 21th Ed. Appleton century croft INC., New York.
- Speck, M.L. (ed.) (1976): Compendium of methods for microbiological examination of food. A.P.H.A., Washington, D.C.
- Thomason, B.M.; Cherry, W.B.; Davis, B.R. and Romales-Lehron, A.A. (1961): Rapid identification of enteropathogenic E.coli in feacal smears by means of flourescent antibody. Ball. Wid. Hlthorg. 25, 117. (cited after El-Bassiony, 1977).
- Tulloch, F.F. Jr.; Ryan, K.J.; Formal, S.S. and Franlin, F.A. (1973): Invasive enteropathogeic E.coli dysentry. An outbreak in 28 students. An Inderna, Med. 79, 1, 13, Abs. of Hyg., 48, 11 (cited after Gadel-Rab, 1983).

## AHMED A-H. AHMED, et al.

# REFERENCES

- Abdel-Rahman, H.A. (1972): Studies on Egyptian white cheese. M.V.Sc. Tkhesis, Fac. of Vet. Med. Cairo Univ.
- Ahmed, A., A-H.; Moustafa, M.K. ad Abdel-Hakiem, E.A. (1987): Sanitary condition of kareish cheese manufactured in Assiut City. Assiut Vet. Med. J. 19 (37): 76-80.
- A.P.H.A. (1978): Standard Methods for the examination of dairy products E.H. Marth (ed), American Public Health Association 14th. Washington D.C.
- Breed, S.R.; Smith, N.R. and Murray, E.G.D. (1957): Bergey's Manual of determinative Bacteriology, 7th Ed., Tindal and Cox Ltd. London.
- Edwards, P.R. and Ewing, W.H. (1972): "Identification of Enterobacteriaceae", 3rd Ed. Burgess Bublishing Company, Minneapolis, Minnesota.
- E1-Bassiony, T.A. (1975): Studies on karish cheese in Upper Egypt M.V.Sc. Thesis Fac. Vet. Med. Assiut University.
- E1-Bassiony, T.A. (1977): Incidence of pathogenic microorganisms and viability of some foodpoisoning bacteria in kareish cheese durig storage. Ph.D. Thesis, Fact. Vet. Med. Assiut University.
- El-Khateib, T.S. (1982): Sanitary condition of sausage in Assiut. M.V.Sc. Thesis, Fac. of Vet. Med. Assiut University.
- Fantasia, L.D.; Mestrandrea, L., Schrade, J.P. Yager, J. (1975): Detection and growth of enteropathogenic E.coli in soft ripened cheese Appl. Microbial 29: 179-185.
- Finegold, S.H. and Martin, W.J. (1982): Baily and Scott Diagnostic microbiology 6th ed. C.V. Mosby Co. St. Louis, Torento, London.
- Frazier, W.C. (1967): "Food Microbiology" 2nd Ed., New York, Mc Graw Hill. Inc.
- Gadel-Rab, H.M. (1983): Enterobacteriaceae in milk and milk prducts. M.V.Sc. Thesis. Fac. Vet. Med. Assiut Univ.
- Ghoneim, N. (1963): Studies on microflora of Damietta cheese. M.V.Sc. Thesis, Cairo Univ. Hall, H.G.; Brown, D.E. and Ewis, K.H. (1967): Examination of market food for coliforms. Appl. Microbiol., 15, 1062.
- Hefnawy, Y. (1980): Studies on sanitary improvement of processed meat (Hamburger) manufactured in Egypt. Ph.D. Thesis. Assiut Univ.
- Hobbs, B.C. and Gilbert, R.J. (eds) (1978): Food poisoning and Food hygiene 4th Ed. English language Book society and Edward Arnold (puplisher) Ltd.
- lordqnov, I.; Slavkov, I. and Bozhilov, B. (1970): Occurrence of (Salmonella in the mammary gland of) ewe. 1st National conference of Salmonellae and salmonellosis in Bulgaria Lgariya 115. Dairy Sci. Abst. 34, 6.
- Kumar, S.; Sinha, B.K. and Sahal, B.N. (1978): Bacterial quality of the raw buffalo milk marketed in and around Patna and its public health importance. Indian J. Dairy Sci., 31, 2: 156.
- Mackie, K.J. and Maccartney, J.E. (1962): Hand book of practical bacteriology 10th Ed. & S. Livingstone Ltd., London.
- Moustafa, T.H.; Kamel, Y.Y.; Ahmed, A.A. and Ismail, A.A. (1975): Studies on the sanitary condition of market milk in Assiut Province, II- The incidence of potentially pathogenic microorgansims. Assiut Vet. 2 (314)181.
- Ridge, L.E. and thomas, M.E.M. (1955): Infection with Providencia type of paracolon bacillus, J. Path. Bact., 69, 335.
- Saudi, A.M.M. (1980): Microbiological studies on food-poisoning microorganisms in some market dairy products. Ph.D. Thesis Fac. Vet. Med. Cairo Univ.
- Shelaih, M.A. (1979): Microbiological studies on Egyptian soft cheese, Ph.D. Thesis, Fac. Vet. Med. Cairo Univ.

The second second second

# The state of the state of

The part of the last of the la