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ENTEROBACTERIACEAE ORGANISMS IN GANDUFLI, STHOMBACAE, CRAB, SHRIMP AND SEPIA FROM ISMAILIA FISH MARKETS

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SUMMARY

Five hundred samples, 25 from each of strombacae, gandufli, shrimp, sepia and crap in each season of the year were collected from Ismailia fish markets. Samples were subjected to bacteriological examinations for the determination of the total Enterobacteriaceae count/ cm^2 surface and/gm muscle and for the isolation and identification of the Enterobacteriaceae organisms.

Average counts of Enterobacteriaceae on the surface of strombacae, gandufli, shrimp, and crab were 10^6, $2x10^5$, $2x10^3$, $<10^2$, 10^2 during Autumn 10^5,$2x10^3$, $3x10^2$, $<10^2$, $<10^2$ during winter 10^7, $2x10^6$, $2x10^4$, $<10^2$ $<10^2$ during spring and 10^9, $4x10^7$, $2x10^5$, $3x10^2$, $<10^2/cm^2$ during summer, respectively.

It could be concluded that the counts of Enterobacteriaceae organisms were higher in strombacae, gandufli, shrimp, sepia then crab and also were higher in summer months than other months of the year.

E.coli, Citrobacter spp., Enterobacter cloacae and Proteus vulgaris were the main isolated organisms.

The hygienic significance of isolated microorganisms was discussed.

INTRODUCTION

Strombacae, gandufli, shrimp, sepia and crab are used for human consumption in many countries of the world. They constitute a very valuable and highly nutritious food where the edible parts are rich in minerals (calcium and phosphorus) and vitamins.

Nowadays shellfish play an important economical role as marine food species due to their exportation to various European countries. On the

other hand, the Molluscs shell contains a high proportion of calcium carbonate where crushed shells are used as supplement to poultry feed (Strou, 1980).

Outbreaks of illness following the consumption of raw partially cooked shellfish were increasingly reported (WHO, 1974; Davies, 1982; Gill et al., 1983; APHA, 1984; Evison, 1985; Guthrie, 1988 and Austin and Austin, 1989). Molluscs are the most significant groups of shell fish associated with gastroenteritis because of their f ilter feeding, which may attract food poisoning microorganisms derived from sewage polluted water and contaminated soil (Bryan, 1980; Banwart, 1981 and Collins and Lyne, 1985).

The present study was planned to assess the public health importance of strombacae, gandufli, shrimp, sepia and carb through the estimation of the Enterobacteriaceae count as well as the isolation and identification of the different members of this group of bacteria.

MATERIAL AND METHODS

Samples:

Samples were collected randomly from Ismailia fish markets for count and isolaton of Enterobacteriaceae organisms from the surface and soft tissue. Such samples included 100 individuals each from Strombcae, Gandufli, shrimp, sepia and crab. Every 25 samples of each group were collected every season the year round . The samples were transferred without delay under aseptic conditions to the laboratory wh they were prepared for bacteriologi examination.

Bacteriological examination:

1-Determination of the Total Enterobacteriac Count:

The technique applied for determination of t total Enterobacteriaceae counts fro surface/cm^2 and muscle/gm was the drop pl method recommended by ICMSF (1978) using violet red bile glucose agar.

II-Isolation and identification Enterobacteriaceae organisms:

The obtained isolates were picked up a streaked onto a slope agar for identificati morpghologically and biochemically.

I) Morphological examination:

Films from pure suspected culture were staine with Gram's method (Jensen's modificatio cited after Cruickshank et al. 1975) a examined microscopically.

II) Biochemical identification:

The isolates were biochemically identified criteria of Edwards and Ewing (1972) a Cruickshank et al. (1975).

The biochemically-identified isolates, whic showed indefinite results, were subjected reidentification by using Entero-tube II fo confirmation.

RESULTS AND DISCUSSION

In the present study 500 samples of strombacae, Gandufli, Shrimp, Sepia and Crab allover one year were examined.

The results from table (1) showed that the average count of Enterobacteriaceae microorganisms were higher in strombacae, Gandufli, shrimp, sepia than crab and this may be due to the extent of pollution (APHA, 1984; Collins and Lune, 1985; West and Coleman, 1986 and Hobbs and Roberts, 1987) or may be from skin, mouth or nose of workers handling the food (Tacher and Clark, 1978). The sources of contamination of molluscs from the view of Bryane, (1980); Carols Abeyta, (1983); APHA, (1984) and National Academy of Sciencas, (1985) may be from sewage, workers, utensils and equipment during processing , distribution and preparation.

The source of contamination of shrimp from the view of Mathana-Saengchindawong (1980); Sunarya et al., (1990); Garnjanagoonchorn and Vibulsresh (1992) and Sunary and Ennatha, (1995) may be from, men flies, during transportation and marketing environmental condition, ants, human hair, small species of water plant, other kinds of crustacea, handling and processing.

The source of contamination of sepia from the view of layer and Varma et al. (1986) and Shrivastova, (1989) may be from aquatic environmental, utensil surface and incubation temperature.

The source of contamination of crab from view of Bullis, (1988) may be from sawage.

The results from table (1) also show that the count of Enterobacteriaceae microorganisms were higher in surface than muscle. These may be due to the protection providing the shell to the flesh from bacterial contamination (Wibowo et al., 1992).

The results from table (1) also show that the count of Enterobacteriaceae microorganisms were higher during summer decreased during spring, followed by autumn then winter and this could be attributed to the seasonal; variation (Chai et al., 1990; Power and Collius, 1990 Martine-Manzanares et al., 1991-b and Puchenkova, 1991).

From the results presented in tables (2&3) it could be concluded that the main isolates were E.coli, Citrobacter spp., Enterobacter cloacae and Proteus vulgaris from strombacae and gandufli with various percentages and this agreed with the findings of Mosa, (1986) and Pucankova, (1991) who examined cultivated molluscs and reached to results nearly similar to these obtained in the present study.

E.coli could be isolated from molluscs by different workers (Mosa, 1986; Paille et al., 1987; Abd El-Massih, 1989 and Colburn et al., 1989).

From the achieved results in table (4) it is evident that the main isolates from shrimp were E.coli, Citiobacter spp., Enterobacter cloacae and Protus vulgaris. The results agree with these reported by

Table (1): Summarized results of Enterobacteriaceae counts in storombacea, Gandaufli shrimp, sepia, and crab :

Samples	Season	Enterobacteriacea Counts					
		Minimum		Maximum		Average	
		Surface	Muscle	Surface	Muscle	Surface	Muscle
Strombacea	Autumn	1×10^5	1×10^3	5×10^7	2×10^4	1×10^6	5×10^3
	Winter	1×10^3	1×10^2	5×10^5	1×10^3	1×10^5	3×10^2
	Spring	1×10^6	5×10^3	7×10^8	2×10^5	1×10^7	4×10^4
	Summer	1×10^7	1×10^4	9×10^9	2×10^6	1×10^9	4×10^5
Gondaufli	Autumn	1×10^4	1×10^2	1×10^6	1×10^3	2×10^5	3×10^2
	Winter	1×10^2	1×10	1×10^4	1×10^2	2×10^3	16.4
	Spring	1×10^5	5×10^2	1×10^7	2×10^4	2×10^6	4×10^3
	Summer	2×10^6	2×10^3	2×10^8	2×10^5	4×10^7	4×10^4
shrimp	Autumn	1×10^2	2×10	1×10^4	2×10^2	2×10^3	2×10
	Winter	1×10	1×10	1×10^2	2×10	3×10	1.2
	Spring	1×10^3	7×10	1×10^5	1×10^3	2×10^4	1×10^2
	Summer	1×10^4	8×10^2	1×10^6	2×10^4	2×10^5	6×10^3
sepia	Autumn	1×10	0	8×10	0	2	0
	Winter	0	0	0	0	0	0
	Spring	5×10	0	1×10^2	0	30	0
	Summer	1×10^2	1×10	1×10^3	6×10	3×10^2	2.8
crab	Autumn	0	0	0	0	0	0
	Winter	0	0	0	0	0	0
	Spring	0	0	0	0	0	0
	Summer	1×10	0	1×10^2	0	4.4	0

n = 25

Table 2: Incidence of isolated Enterobacteriaceae organisms from strombacae:

Isolates	Autumn				Winter				Spring				Summer			
	Surface		Muscle		Surface		Muscle		Surface		Muscle		Surface		Muscle	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
E. coli	13	52	8	32	10	40	5	20	15	60	10	40	20	80	15	60
citrobacter spp.	6	24	1	4	5	20	0	0	7	28	2	8	9	36	4	16
Enterobater cloacae	7	28	2	8	6	24	1	4	8	32	3	12	10	40	5	20
proteus vulgaris	8	32	3	12	7	28	2	8	9	36	4	16	12	48	6	24

No. = Number of the positive samples to the microorganism.

Table 3: Incidence of isolated Enterobacteriaceae organisms from Gando fli:

Isolates	Autumn				Winter				Spring				Summer			
	Surface		Muscle		Surface		Muscle		Surface		Muscle		Surface		Muscle	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
E. coli	12	48	7	28	9	36	4	16	14	56	9	36	19	76	14	56
citrobacter spp.	5	20	0	0	4	16	0	0	6	24	1	4	8	32	3	12
Enterobater cloacae	6	24	1	4	5	20	0	0	7	28	2	8	9	36	4	16
proteus vulgaris	7	28	2	8	6	24	1	4	8	32	3	12	11	44	5	20

No. = Number of the positive samples to the microorganism.

Table 4: Incidence of isolated Enterobacteriaceae organisms from shrimp :

Isolates	Autumn				Winter				Spring				Summer			
	Surface		Muscle		Surface		Muscle		Surface		Muscle		Surface		Muscle	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
E. coli	5	20	2	8	3	12	1	4	6	24	2	8	9	36	4	16
citrobacter spp.	2	8	0	0	1	4	0	0	3	12	0	0	6	24	1	4
Enterobater cloacae	6	24	2	8	5	20	1	4	6	24	2	8	7	28	3	12
proteus vulgaris	0	0	0	0	0	0	0	0	0	0	0	0	6	24	1	4

No. = Number of the positive samples to the microorganism.

Table 5: Incidence of isolated Enterobacteriaceae organisms from sepia :

Isolates	Autumn				Winter				Spring				Summer			
	Surface		Muscle		Surface		Muscle		Surface		Muscle		Surface		Muscle	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
E. coli	2	8	0	0	0	0	0	0	3	12	0	0	8	32	3	12
citrobacter spp.	0	0	0	0	0	0	0	0	0	0	0	0	1	4	0	0
Enterobater cloacae	4	16	0	0	0	0	0	0	5	20	0	0	7	28	2	8
proteus vulgaris	0	0	0	0	0	0	0	0	0	0	0	0	6	24	1	4

No. = Number of the positive samples to the microorganism.

Table 6: Incidence of isolated Enterobacteriaceae organisms from crab :

Isolates	Autumn				Winter				Spring				Summer			
	Surface		Muscle		Surface		Muscle		Surface		Muscle		Surface		Muscle	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
E. coli	0	0	0	0	0	0	0	0	0	0	0	0	2	8	0	0
citrobacter spp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Enterobater cloacae	0	0	0	0	0	0	0	0	0	0	0	0	1	4	0	0
proteus vulgaris	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

No. = Number of the positive samples to the microorganism.

Karunasagar et al., (1992) who isolated enterobacter and coliforms from shrimp, and with these achieved by Xu et al., (1992), who isolated Protus vulgaris from shrimp and agree also with Singh and Kulshreshe (1994) isolated E. coli, but not in agreement with the results obtained by Garnjanagoonchorn and Vibulsresh (1992), Mohney et al. (1992), Peranginangin et al. (1992) and Sunarya and Ennatha (1995) who failed detection of E.coli.

Escherichia coli and Enterobacter cloacae could be isolated from crab (Table 6) and these results agreed with the findings of Bullis edt al., (1988) and Ho et al. (1994).

Bryan, (1980) reported that sea foods w implicated as vehicles in approximately 11% food borne disease outbreak in the Untied St while molluscs were involved in about 1.9%

The problem with shellfish is that the potenti harmful bacteria they carry may readily mult to the point where they can actually cause ha Thorough cooking of shellfish make them for human consumption (Gill et al., 1983; Gutl 1988 and Vernam and Evan , 1991).

Jennings, (1975), reported that shelfish responsible for food-borne outbreaks and ad that food-borne illness was mainly due Escherichia coli microorganisms was associ

with outbreaks of gastrointestinal disease (Delopne, 1903), coli-enteritis in children, and peritonitis, meningitis, cystitis, pyelonephritis, appendicitis and ottis in adults (Pyatkins and Krivoshein, 1980). Food-borne outbreaks from shellfish was due to members genera proteus, which have been incriminated in cases of gastroenteritis (Cooper et al., 1941, and Cherry et al., 1946), summmer diarrhea in infants and urinary tract infections (I.C.M.S.F, 1978) food poisoning (Frazier, 1967 and Halstead, 1967). Food borne outbreaks from shellfish were also due to Citrobacter spp. Jennings (1975) concluded that many kinds of frozen seafood were the vehicle.

Reported of W.H.O., (1974) stated that molluscs constitute a risk to human as they may cause enteric disease including typhoid and paratyphoid fever, in addition hepatitis A and cholera were reported by Bryan, (1980).

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