

MICROBIAL HAZARD OF SALTED OM EL-KHOLOUL (WEDGE SHELL-DONAX TRUNCULUS)

By

**NADA K. MANSOUR, NABIL A. YASSIEN, HEMAT M. IBRAHIM*
and MOHAMED K. ELMOSSALAMI**

Dept. of Food Hygiene, Fac. of Vet. Med., Cairo Univ.

* Dept. of Food Hygiene, Fac. Vet. Med., Moshtohor, Benha Univ.

SUMMARY

Sixty samples of salted Om El-Kholoul were collected from different localities in Giza, Alexandria and Ismailia. Collected samples were examined organoleptically and subjected to bacteriological examination for enumeration of Aerobic, enterobacteriaceae, Staph. aureus, enterococci counts as well as isolation and identification of *Vibrio parahaemolyticus*.

The mean counts/gm of Aerobes, Enterobacteriaceae, Staph. aureus, Strept. faecalis and Strept. faecum were 4.9×10^5 , 4.8×10^4 , 1.9×10^5 , 1.5×10^5 , and 8×10^2 , respectively. *V. parahaemolyticus* could be isolated from all examined samples.

The weight, pH and sodium chloride percent of ten samples were estimated and the inhibitory effect of lemon juice (*Citrus aurantifolia*) on the microbial load of such samples was noticed.

The public health significance of isolated microorganisms and the suggestive measures for improvement of the microbial quality of the product were discussed.

INTRODUCTION

Molluscan shell fish are normally found in water near the shore and estuaries, they are subjected to contamination of run off water carrying soil microorganisms and sewage outfall.

Nowadays, bivalved molluscus are used for human consumption all over the world, as they constitute a very valuable and highly nutritious in all nutrients as protein, calcium, phosphorus and vitamins (Waterman, 1980).

Om El-Kholoul (Wedge shell-*Donax trunculus*) is bivalved molluscus, collected from marine coasts, cleaned from dirt and planktons, then salted by

addition of sodium chloride. The product is consumed after elapse of 48 hours from salting, mainly by pupils, children and picnickers.

Harvesting of bivalved molluscus from faecal polluted areas constitutes a major public health hazard (Bryan, 1980; Banwant, 1981; Gill et al; 1983 Evison, 1985). Many pathogenic and toxigenic bacteria could be concentrated in the molluscus as they act as filter feeders (APHA, 1984).

Abd El-Massih (1989) mentioned that molluscan shell fish harvested from water of El-Max coast (Alexandria) were heavily polluted by *E. coli* as compared with those harvested from Ismailia and edco coasts. Such variations were attributed to the degree of sewage pollution.

Sea food products were incriminated in many events of food-borne illness, which were due to *Staphylococci*, *Streptococci*, *Clostridia*, *Salmonellae* and *Vibrio* species (Carlos, 1983, 1983; Evison, 1985, Desenclos, 1991).

Molluscus may also harbour chemical residues, parasites and viruses (Okazaki & Panietz, 1981; Imam et al., 1992; Pontenfact et al., 1993).

The present investigation was carried out to evaluate the bacteriological quality and sanitary condition of Om El-Kholoul harvested from different shores and to investigate the effect of lemon juice (*Citrus aurantifolia*) on such quality.

MATERIAL AND METHODS

Sixty samples of Om El-Koloul were collected from different localities in Giza, Alexandria, Ismailia in sterile polyethylene bags and transported without delay to the laboratory in a sampling box, where the edible part of six samples were subjected to the following investigations:

- 1- Organoleptic examination.
- 2- Determination of aerobic, Enterobacteriaceae and *Staphylococcus aureus* counts/g according to (ICMSF, 1978).
- 3- Determination of Enterococci count using Enterococcus Selective Differential medium (ESD) according to Efthymious et al. (1974).
- 4- Isolation and identification of *V. parahaemolyticus* using Tiosulphate Citrate Bile salt Sucrose media (TCBS) according to APHA (1984).

The weight, pH-value using the digital pH-meter (Hofmann, 1987) and sodium chloride percent according to AOAC (1990) were determined in ten out of the above samples.

The inhibitory effect of lemon juice (*Citrus aurantifolia*) on the above mentioned bacterial groups and on the viability of *V. parahaemolyticus* was also studied by adding 0.5ml of the natural lemon juice to 2.5 gm of the edible part of Om El-Kholoul for 30 seconds.

RESULTS and DISCUSSION

Table (1): Average count of microbial load in Om El- Kholoul.

Count	Minimum	Maximum	Mean	St. error \pm
APC	3×10^3	3×10^6	4.9×10^5	94.01
Enterobacteriaceae	2×10^3	2×10^5	4.8×10^4	24.84
Staph. aureus	$\leq 10^2$		1.9×10^5	1.2×10^5
<u>Enterococci</u>				
E. faecalis	$\leq 10^2$	6.0×10^6	1.5×10^5	1.2×10^5
E. faecium	$\leq 10^2$	2×10^4	8×10^2	0.14
E. intermediate	$\leq 10^2$	$\leq 10^2$	0.0	0.0

Organoleptic examination of Om El-Kholoul revealed that the colour of the internal contents was creamy with greyish centre, orange or yellow with darker centre with the presence of some characteristic salty fluid (Figure 1). Contents filled fast all shell in most of samples, but in some ones the contents were shrunk with dirty greyish colour with unpleasant odour and salty taste.

It is obvious from table (1) that the mean counts of Aerobes, Enterobacteriaceae and Staph. aureus were 4.9×10^5 , 4.8×10^4 and 1.9×10^5 /gm respectively. These findings nearly agreed with those findings nearly agreed with those reported by Abd El-Aziz (1991). While lower results were obtained by Abd El-Rahman et al. (1994). In this respect, Bryan (1970) reported that fish and shell fish products were responsible for 5.7% of

reported outbreaks of Staph. aureus. In Egypt, the hazards increase also because of the high number of Staph. aureus/gm, besides its harvesting from polluted areas as well as its nature as filter feeders. Moreover hazards are also expected from the consumption of Om El-Kholoul due to the higher pH value of the product (good environment for growth of microbes).

Dealing with Enterococci, enterococcus faecalis had the highest count/gm (1.5×10^5) followed by E. faecium (8×10^2), while E. intermediate could not be isolated. Such counts were higher than those found by Ibrahim (1996). The Enterococci have a distinctive role as indicator for poor sanitation. Strept. faecalis can grow at a wide range of temperature with standing heat treatment,

Table (2): Effect of lemon juice on bacterial associations of *Om El- Kholoul*.

Sample No.	Weight		pH	Salt content %	Untreated		Treated	
	Total	Contents			%	AP count	Enterobact. count	AP count
1	0.638	0.225	6.94	9.65	2x10 ⁴	1x10 ⁴	10 ³	≤ 10 ²
2	0.770	0.216	7.15	9.95	3x10 ⁴	10 ⁴	2x10 ⁴	6x10 ³
3	0.778	0.311	7.04	8.78	10 ⁵	9x10 ³	6x10 ³	≤ 10 ²
4	0.676	0.263	7.10	8.19	10 ⁴	10 ⁴	4x10 ³	2x10 ³
5	0.648	0.238	7.10	9.07	3x10 ³	2x10 ³	2x10 ²	≤ 10 ²
6	1.074	0.415	7.10	5.84	3x10 ⁶	2x10 ⁵	3x10 ⁴	≤ 10 ²
7	1.330	0.457	7.14	5.85	9x10 ⁵	10 ⁵	10 ⁴	≤ 10 ²
8	0.909	0.306	6.80	9.65	2x10 ⁵	2x10 ⁴	8x10 ³	≤ 10 ²
9	1.225	0.490	7.02	7.61	5x10 ⁴	2x10 ⁴	6x10 ³	≤ 10 ²
10	1.289	0.416	7.11	3.80	6x10 ⁵	10 ⁵	5x10 ³	≤ 10 ²
Mean	0.934	0.337	7.05	7.81	4.9x10 ⁵	4.8x10 ⁴	8.5x10 ³	8.6x10 ²
±S.E.	± 0.050	± 0.307	± 0.0319	± 0.649	± 94.009	± 24.836	± 9.546	± 4.251

AP= Aerobic plate count

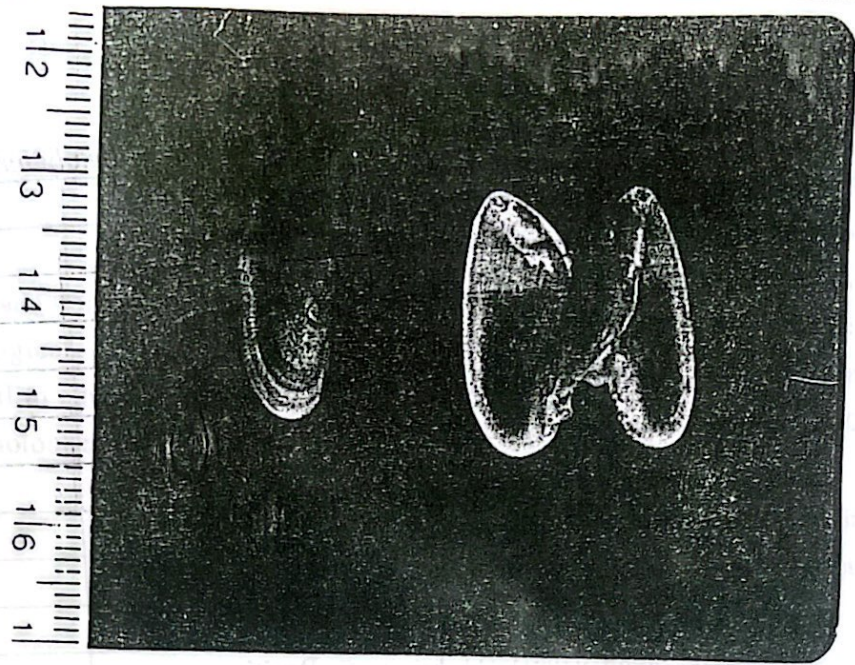


Figure (1): Om El- Kholoul (Wedge shell - *Donax trunculus*).

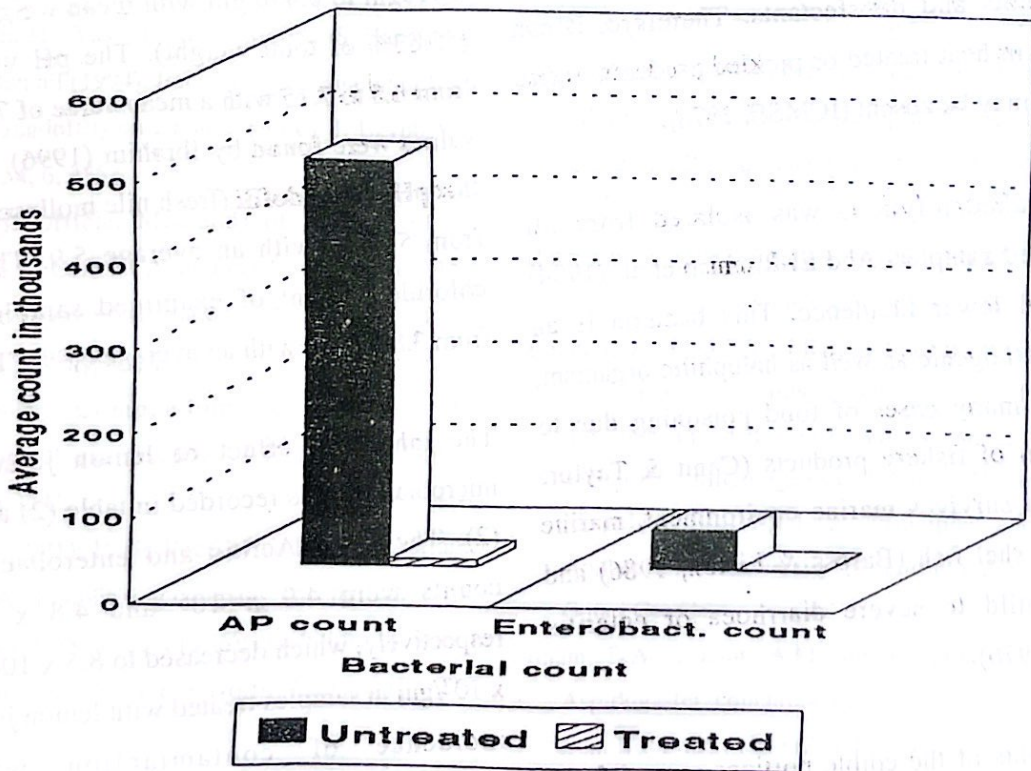


Figure (2): Effect of lemon juice on Aerobic and Enterobacteriaceae count.

Table (3): Effect of lemon juice on *Vibrio parahaemolyticus* of Om El- Kholoul.

Sample No.	Untreated samples	Treated samples
1	Positive	negative
2	Positive	Positive
3	Positive	negative
4	Positive	Positive
5	Positive	negative
6	Positive	negative
7	Positive	negative
8	Positive	negative
9	Positive	negative
10	Positive	negative
No. / %	10/100	2/20

tolerating sodium chloride, low pH, drying, detergents and disinfectants. Therefore, it can persist in heat treated or pickled products, where *E. coli* may be absent (ICMSF, 1978).

V. parahaemolyticus was isolated from all examined samples. Abd El-Rahman et al. (1994) reported lower incidence. This bacteria is an enteropathogenic as well as halophilic organism, causing many cases of food poisoning due to ingestion of fishery products (Cann & Taylor, 1981). It survives marine environment, marine fish and shell fish (Baross & Liston, 1986) and causes mild to severe diarrhoea or enteritis (WHO, 1976).

The weights of the edible portions ranged from

0.216gm to 0.490 gm with mean weight 0.337 (35.83% of total weight). The pH value varied from 6.8 to 7.15 with a mean value of 7.05. Low values were found by Ibrahim (1996) who stated that pH of Gandoffli (fresh Nile molluscan) ranged from 5.2-6.3 with an average 5.9. The sodium chloride percent of examined samples ranged from 3.8-9.95% with an average 6.9% (Table 2).

The inhibitory effect of lemon juice on microbial loads is recorded in table (2) and figure (2). The mean Aerobic and enterobacteriaceae counts were 4.9×10^5 and 4.8×10^4 /g respectively, which decreased to 8.5×10^3 and 6×10^2 /gm in samples treated with lemon juice. The incidence of contamination with *V. parahaemolyticus* reduced in examined samples.

from 100% to 20% after being treated with lemon juice.

It is evident therefore that addition of lemon juice improved the bacteriological quality of the product. Thus, it is fruitful to advice addition of lemon juice to Om El-Kholoul to safeguard the health of consumer.

REFERENCES

- Abd El-Aziz, A.H. (1991): Microbial studies on some marketed shell fishes. M.V.Sc. Fac. Vet. Med., Moshtohor, Benha Univ.
- Abd El-Massih, S.G. (1989): Occurrence of food poisoning agents in common molluscan shell fish in Alexandria. M.V.Sc. Alexandria Univ.
- Abd el-Rahman, H.; Yassien, M.A.; Mervat, S. Hanafi and Takwa, H. Ismail (1994): Bacteriological quality of fresh molluscs (*Gandoffli*) on Ismailia shores. J. Egypt. Vet. Med. Ass., 54, 6, 485.
- AOAC (1990): Official Methods of Analysis of the Association of Official Analytical Chemists. 864. 15th ed. published by the Association of Analytical Chemists, Inc. Arlington, Virginia, USA.
- APHA (1984): Compendium of Methods for Microbiological examination of Foods. 2nd Edition, Washington, D.C.
- Banwart, G.J. (1981): Basic Food Microbiology, Abridged Text book AVI publishing company. Inc, pp 123.
- Baross, J. and Liston, J. (1986): Isolation of *Vibrio parahaemolyticus* from the North-West Pacific 217, 1263.
- Bryan, F.L. (1970): The epidemiology of Staphylococcal food poisoning N.Y. Agr. exp. Sta, Geneva, Res. Circ.
- Bryan, F.L. (1980): epidemiology of food-borne disease transmitted by fish, shell fish and marine crustaceans in the United States, 1970-1978. J. Food Prot. 43, 859.
- Cann, D.C. and Taylor, L.L. (1980): A study of the incidence of *Vibrio parahaemolyticus* in Malaysian shrimps undergoing processing for export J. Hyg. Camb. 87, 485.
- Carlos Abeyta, J.R. (1983): Bacteriological quality of fresh seafood products from Seattle Retail markets. J. Food Prot., 46, 901.
- Desenclos, J.C. (1991): *Vibrio* illness in the Florida raw oyster eating population. Am. J. Epidemiol., 134, 290.
- Efthymious, C.; Baccash, P.; Labmbard, V. and Epstein, D. (1974): Improved isolation and differentiation of Enterococci in cheese. J. Applied Microbiol., 28, 417.
- Evison, L.M. (1985): Bacterial pollution of coastal waters in the U.K. and Mediterranean. J. Applied Bacteriol. Sym. Suppl. pp. 815.
- Gill, O.N.; Cubitt, W.D.; Mc Swiggan, D.A.; Watney, B.M. and Barlett, C.L.R. (1983): Epidemic gastroenteritis caused by SRSV contaminated oysters. Br. Med. J. 287, 1532.
- Hoffmann, K. (1987): Der pH wert ein Qualitates Kriterium fuer Fleisch. Fleisch Fleischwirtsch. 67, 557.
- Ibrahim, A.M. (1995): Microbiological studies on Nile bivalvia "*Anodonta rubens*". 4th Sci. Cong. Proc., April 3-6, 1996, Vet Med. J., Giza. Vol. 44, No. 2, 189.
- ICMSF (1978): Microorganisms In Foods, II-Sampling For Microbiological Analysis. Principles and Specific Applications. University of Toronto press. Toronto, Ontario Canada.
- Imam, E.A.; Nassar, A.M. and Ibrahim, A.M. (1992): *Aspidogaster conchiola* (Trematoda; Aspidogastrea), from fresh water clam (*Anodonta rubens*). J. Egypt. Vet. Med. Ass., 52, 431.

Okazaki, R.K. and Panietz, M.H. (1981): Depuration of twelve trace metals in tissues of the oysters *C. gigas* and *C. virginica*. *Marine biol.*, 63., 113.

Pontenfact, R.; Bishai, F.; Hockin, J.; Bergeron, G. and Parent, R. (1993): Norwalk viruses associated with a gastroenteritis outbreak following oyster consumption. *J. Food Prot.*, 56, 604.

Waterman, J.J. (1980): **Processing mussels, cockles and whelks.** Ministry of agriculture, fisheries and Torry, Research station. Torry advisory note No. 12

WHO (1976): **Microbiological aspects of food hygiene.** Technical Report Series No. 598pp. 21-23, Geneva, Switzerland.

REFERENCES

Waterman, J.J. (1980): **Processing mussels, cockles and whelks.** Ministry of agriculture, fisheries and Torry, Research station. Torry advisory note No. 12

WHO (1976): **Microbiological aspects of food hygiene.** Technical Report Series No. 598pp. 21-23, Geneva, Switzerland.

Okazaki, R.K. and Panietz, M.H. (1981): Depuration of twelve trace metals in tissues of the oysters *C. gigas* and *C. virginica*. *Marine biol.*, 63., 113.

Pontenfact, R.; Bishai, F.; Hockin, J.; Bergeron, G. and Parent, R. (1993): Norwalk viruses associated with a gastroenteritis outbreak following oyster consumption. *J. Food Prot.*, 56, 604.

Waterman, J.J. (1980): **Processing mussels, cockles and whelks.** Ministry of agriculture, fisheries and Torry, Research station. Torry advisory note No. 12

WHO (1976): **Microbiological aspects of food hygiene.** Technical Report Series No. 598pp. 21-23, Geneva, Switzerland.