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CENTRAL MARINE FISHERIES RESEARCH INSTITUTE  
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## 86. A STUDY ON THE BACTERIAL QUALITY OF BROWN MUSSEL *PERNA INDICA* AND ITS PURIFICATION

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### ABSTRACT

Brown mussel (*Perna indica*) samples were collected periodically during 1983-1985 from Vizhinjam Central Marine Fisheries Research Institute farm and also from the natural beds and were studied for their bacterial quality. The seawater samples surrounding the mussels were also collected along with the mussel samples and analysed for physical, chemical and bacteriological qualities. The mussel samples were subjected to purification by employing different purification methods. The total bacterial count of cultured brown mussels and natural bed brown mussels ranged between  $10^2$  to  $10^3$  organisms per ml of mussel fluid. The T. B. C. of the sea water around cultured brown mussels and natural bed brown mussels ranged between  $10^2$  to  $10^3$  organisms per ml of seawater. The faecal coliforms were found to be very low and they were in permissible limits. The pathogenic bacteria *Salmonella*, *Streptococci* and *Staphylococci* were absent. The variations in pH, temperature, salinity and dissolved oxygen of the seawater samples were insignificant. The mussels were subjected for purification by employing different purification methods and chlorination was found to be better.

### INTRODUCTION

The increasing pollution in seawater poses threat to mussel farming and the discharge of industrial effluents and sewage in these waters necessitates purification of mussels before marketing because the shell fish are filter feeders and can concentrate pathogenic bacteria that may be present in the surrounding waters. Fraiser et al (1984) have reported the incidence of *Salmonella* in clams and oysters in Florida. Deapola et al (1983) have reported the occurrence of various strains of *Vibrio cholerae* in shell fish, sediments and water along the US Gulf coast. Though information on the qualitative and quantitative aspects of bacterial flora is available on fish and also offshore waters, data in respect of shell fish in the tropical seas are scanty.

Durairaj et al (1983) have studied the bacteria flora of edible oysters of Tuticorin waters during the year 1982-83. The present paper deals with the bacterial flora of the mussel samples collected from Vizhinjam. Attempts were also made to purify the mussels employing different purification methods.

### MATERIAL AND METHODS

Fresh brown mussels (*P. indica*) were collected at regular intervals from Central Marine Fisheries Research Institute from the natural bed which exist in the vicinity of Vizhinjam Bay. Seawater samples were also collected on few occasions. The samples were collected in sterile containers and brought to Tuticorin in live condition.

A sample of 2 to 5 mussels were used for preparing mussel fluid which was prepared by blending with equal amount by weight of mussel with equal volume of sterile 0.85% saline solution and used as dilution for plating purpose. Plate count of the mussel liquid and seawater samples were determined by preparing duplicate plates, tryptone Glucose Yeast extract agar. The plates were incubated at 37°C for 48 h (Presnell and Kelly 1961) and APHA (1976). Ec broth with incubation at 44.5°C and Eosin Methulene Blue Agar were used for the enumeration of faecal coliforms (APHA 1976) and for *E. coli* Tergital 7 Agar were used. For enumeration of coagulase + Ve staphylococci Baird parker agar and for faecal streptococci KF Agar were employed. For the enumeration of *Salmonella*, Brilliant Green agar, Bismuth sulphite agar and Triple sugar Iron Agar were used. During the period under report farmwater samples, and natural bed seawater were collected along with the mussel samples. The hydrological parameters of the seawater samples such as dissolved oxygen, salinity, pH were estimated on the spot according to the standard methods of water analysis (AOAC 1970).

The mussel samples were subjected to purification by employing different purification techniques. Various authors have described different purification methods for shell fishes. Eyles and Davy (1984) have described purification methods for commercial depuration of shell fish. The purification plant was of the recirculating type employing UV light for sterilisation. Balachandran et al (1984) suggested the method of chlorination upto 5 ppm at the end of depuration to improve the bacterial quality of shellfishes. They have suggested the depuration of mussels in seawater for a period of 16-18 h. Ray (1984) has suggested a depuration period of 36 to 48 h for purification of oysters. Three methods were followed during the recent study. The mussel samples were thoroughly washed and allowed to remain in filtered seawater for 24 hours with change of water once in 12 h. The mussel samples were kept for 24 h and 48 h in filtered and aerated seawater with the change of water once in 12 h. (3) In the 3rd method the depurated samples were kept in filtered seawater and chlorinated at 2, 2.5 and 3ppm for 3 h and then taken out and thoroughly washed and examined for bacterial quality.

TABLE 1. *Bacteriological studies of mussels and farm water collected from Central Marine Fisheries Research Institute farm at Vizhinjam*

Date	Farm Mussels		Farm Seawater	
	TBC/g	Faecal coli/100g	TBC/ml	Faecal coliform/100ml
1. 23.6.83	23X 10 <sup>3</sup>	12	14.8X 10 <sup>3</sup>	6
2. 26.7.83	32.25 X 10 <sup>3</sup>	14	14.0X 10 <sup>2</sup>	8
3. 25.8.83	12.25 X 10 <sup>2</sup>	Nil	6.5X 10 <sup>2</sup>	Nil
4. 27.9.83	9.55 X 10 <sup>3</sup>	16	9.4X 10 <sup>2</sup>	12
5. 25.10.83	11.56 X 10 <sup>2</sup>	12	3.75 X 10 <sup>2</sup>	6
6. 15.11.83	11.56 X 10 <sup>2</sup>	12	4.4 X 10 <sup>2</sup>	8
7. 21.5.84	16 X 10 <sup>2</sup>	Nil	3 X 10 <sup>2</sup>	Nil
8. 20.7.84	12.4 X 10 <sup>2</sup>	Nil	4.2 X 10 <sup>2</sup>	Nil
9. 17.8.84	14.5 X 10 <sup>2</sup>	Nil	3.4 X 10 <sup>2</sup>	Nil
10. 26.10.84	0.5 X 10 <sup>3</sup>	Nil	11.8 X 10 <sup>2</sup>	Nil
11. 29.11.84	2.7 X 10 <sup>3</sup>	Nil	2.9 X 10 <sup>2</sup>	Nil
12. 23.1.85	1.6 X 10 <sup>3</sup>	Nil	3.7 X 10 <sup>2</sup>	Nil
13. 27.3.85	8.7 X 10 <sup>2</sup>	Nil	4.8 X 10 <sup>2</sup>	Nil
14. 25.4.85	9.4 X 10 <sup>2</sup>	7	4.4 X 10 <sup>2</sup>	6 [Natural bed Mussel & seawater)
15. 26.6.85	8.0 X 10 <sup>2</sup>	14	3.2 X 10 <sup>2</sup>	8 "

## RESULTS AND DISCUSSION

### *Bacterial flora of mussels and seawater samples*

Table 1 indicates that the total viable count of the mussels ranged from  $8.51 \times 10^2$  to  $32.25 \times 10^3$  and the TBC of the farm water ranged from  $2.9 \times 10^2$  to  $14.8 \times 10^3$ /ml. The TBC of the natural bed mussels ranged from  $8.0 \times 10^2$  to  $9.4 \times 10^2$ /g and the TBC of the natural bed mussels ranged from  $8.0 \times 10^2$  to  $9.4 \times 10^2$ /g and the TBC of the farm water ranged from  $3.2 \times 10^2$  to  $4.4 \times 10^2$ /ml. The TBC of the surrounding seawater was consistently lower than the corresponding counts of the mussels. This is in agreement with the findings of Durairaj et al (1983). The TBC of the seawater and natural bed mussels was found to be lower than that of the farm mussels. This may be due to the obvious reason that the Vizhinjam bay is subjected to sewage and domestic pollution. This is in agreement with the findings of Thangappan Pillai (1980).

The faecal coliforms in the farm mussel samples and farm seawater sample ranged from

nil to 16/100/g and nil to 12/100/cc respectively. The faecal coliforms in the natural bed mussel and seawater ranged from 7 to 14/100/g and 6 to 8/100 ml respectively. According to APHA (1970) the permissible limits of coliforms is 230/100 g in depurated oysters. Coliform counts were reported to be maximum under low salinity conditions (Presnell and Kelly 1961). From the observations given in Table 1 it is observed that highest counts of coliform (16 Nos/100 gm) was noticed in mussels during September '83 when lower salinity of 30.9 ppt was recorded. David Hussong et al (1981) has indicated that coliform counts of oysters were found to increase during higher temperature. But no such correlation was observed at Vizhinjam by the authors. The other pathogens like *Salmonella*, *Streptococci*, *Staphylococci* and *E. coli* were absent in these waters.

### *Hydrological parameters of seawater samples*

The data of the hydrological parameters is given in Table 2. There was no marked fluctuations in pH and other hydrological parameters.

TABLE 2. *Hydrological parameters of seawater samples collected at Vizhinjam*

Period of study	Temperature °C	Dissolved Oxygen ppl	pH	Salinity ppt
1. 23.6.83	25.2	6.2	8.4	36.9
2. 26.7.83	25.0	5.6	8.3	32.5
3. 25.8.83	23.5	5.2	8.2	33.0
4. 27.9.83	25.0	5.4	8.5	30.9
5. 25.10.83	20.0	5.0	8.3	35.9
6. 15.11.83	28.6	5.2	8.5	35.9
7. 21.5.84	27.6	5.4	8.5	35.9
8. 20.7.84	25.0	4.8	8.1	33.5
9. 17.8.84	25.0	4.6	8.7	33.4
10. 26.10.84	26.0	5.0	8.3	36.5
11. 29.11.84	28.4	5.8	8.5	34.7
12. 23.1.85	28.1	6.0	8.4	35.9
13. 27.3.85	27.0	6.0	8.4	37.0
14. 24.4.85	30.5	4.2	8.4	38.8*
15. 26.6.85	28.0	3.2	8.5	31.0*

\* Natural bed seawater samples.

TABLE 3. *Purification studies of mussels collected from Vizhinjam*

Particulars	29.11.84	27.3.85
1. Raw mussel	$2.7 \times 10^3$	$8.7 \times 10^2$
2. Seawater sample	$2.9 \times 10^2$	$4.8 \times 10^2$
3. Filtered sea water	90/ml	85/ml
4. Mussel samples after washing thoroughly in seawater for 24 h	$2.2 \times 10^3$	$7.5 \times 10^2$
5. Mussel sample after 24 h aeration in filtered seawater	$2.4 \times 10^2$	$6.0 \times 10^2$
6. Mussel sample after 48 h aeration with change of filtered seawater	$2.2 \times 10^2$	$6.0 \times 10^2$
7. The depurated mussel samples after chlorination for 3 h and then thoroughly washed.		
1. 2 ppm	$2.4 \times 10^2$	$2.9 \times 10^2$
2. 2.5 ppm	$1.3 \times 10^2$	$2.4 \times 10^2$
3. 3 ppm	$2.0 \times 10^2$	$2.0 \times 10^2$

#### *Purification of mussels*

From the data given in Table 3 it was noted that there was reduction in bacterial load as a result of purification by the methods already described. Mahadevan (1980) has suggested depuration of oysters for 24 h in seawater, and then chlorination with 3 ppm for 1 h for better quality. Balachandran et al (1984) have suggested chlorination upto 5 ppm after the depuration to improve the bacterial quality. Ray (1984) is of the view that chlorination is less attractive when compared with the other forms of purification. The observations recorded by the authors (Table 3) indicate that the bacterial load of the mussels could be brought down effectively, either by washing in the filtered seawater for 24 h (average reduction 14.4%) or by keeping them in aerated seawater for 48 h (average reduction 58.2%). The bacterial quality could be further improved by chlorination at 3 ppm at the end of depuration (average reduction 84.5%). As the initial TBC load of the mussels in Vizhinjam is well below the permissible limit, the purification of mussels could be done either by keeping the mussels in filtered aerated seawater upto 48 h with the change of water once in 13 h or by chlorination at 3 ppm level at the

end of depuration in filtered, seawater. However the chlorination gives better results. The Chlorinated mussels were thoroughly washed for marketing purpose.

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