

## **Bacterial Counts in Two Species (*Scomberomorus juttatus* and *Otolithes ruber*) of Fresh South-Harvested Fish, While Loading in Kazeroon**

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**Abstract:** Since fish meat which has a wide range of consumption among people may harbor pathogens its microbial quality is crucially important in public health. This study focuses on the level of bacterial counts in two kinds of fish (*Scomberomorus juttatus*, *Otolithes ruber*) harvested from some South coastal areas of Iran (Booshehr Province) subjected to refrigerated-transportation to Kazeroon. Hence, this research was commenced to address whether freshly harvested fish stored under 2-3 h refrigeration presented any likely problems such as generating bacterial levels in excess of regulatory standards. In this study, 25 samples of *Scomberomorus juttatus* and 25 samples of *Otolithes ruber* were studied for total bacteria, coliform and *V. parahaemolyticus* counts. However, none of the samples exceeded the standard value and this study shows that the overall microbial quality of fresh South-harvested fish is acceptable and according to the standard values, while it is loaded in Kazeroon.

**Key words:** Total bacterial count, coliform, *Vibrio parahaemolyticus*, fish, Kazeroon

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### **INTRODUCTION**

The importance of fish as a source of animal protein is well understood and the avoidance of quality losses and the contamination of harvested fish with pathogenic bacteria are very important. The number and the nature of the fish bacteria are influenced by many factors such as sea water pollution, temperature, method of capture, preservation methods and handling practices.

Numerous species of fish occur along the Iranian coastline but this study focuses on two main species which have a high consumption in Kazeroon. These fish species are harvested at several stations along the coasts of the Persian-Gulf in Iran and are transported at ambient temperatures (approximately 0°C) prior to loading and marketing in Kazeroon. Transportation usually takes 2-3 h and this study was conducted to determine the effect of this transportation period on the bacteriological quality of the fish harvested from this South-coastal area of Iran.

### **MATERIALS AND METHODS**

Because of being carried in refrigerated transporting vehicles, it usually took the harvested fish 2-3 h to reach the markets in Kazeroon. In these vehicles, fish were carried in ice-powder in order to keep the temperature

around 0°C. In this study, 25 samples of *Scomberomorus juttatus* and 25 samples of *Otolithes ruber* were collected randomly, while the fish were being loaded in Kazeroon (5-7 o'clock in the morning). Samples were collected in an ice box and taken to the laboratory where they were immediately examined duplicately for the bacteriological quality. Fish were analyzed at three different treatment levels: (a) skin, (b) muscle and (c) gills.

Samples of different parts (skin, muscle and gills) of every fish were prepared for analysis according to the Iranian Institute of Standard and Industrial Studies (2006) and examined quantitatively for standard plate count, coliforms and *V. parahaemolyticus*.

Standard plate count was performed according to the Iranian Institute of Standard and Industrial Studies (2008), Coliforms and *V. parahaemolyticus* were enumerated using the techniques specified in the Iranian Institute of Standard and Industrial Studies (2005, 2007), respectively. All bacteriological media were obtained from Merck, Germany. SPSS software (Repeated measure and t-test) were employed to statistically evaluate the data.

### **RESULTS AND DISCUSSION**

Microbiological standards for fish are prescribed in the Iranian Institute of Standard and Industrial Studies

(1999). It stipulates that the total content of the fish shall not contain a total mesophilic bacteria, faecal coliform and *Vibrio parahaemolyticus* density of more than  $10^6-10^7$ ,  $4.0 \times 10^2$  and  $1.0 \times 10^2$  colony forming units (cfu)  $g^{-1}$ , respectively.

Out of total 50 samples, 5 skin samples (10%) and 6 gill samples (12%) exceeded the standard value of total mesophilic aerobic count which is  $10^6-10^7$  cfu  $g^{-1}$ . In case of total coliform count, 1 skin samples (2%) and 2 gill sample (4%) crossed the standard value which is  $4 \times 10^2$  cfu  $g^{-1}$ .

Similarly, 2 skin samples (4%) and 1 gill sample (2%) were found crossing the standard value of *Vibrio parahaemolyticus* count ( $10^2$  cfu  $g^{-1}$ ). The average counts for total mesophilic aerobic bacteria, coliform and *Vibrio parahaemolyticus* were found to be  $2.8 \times 10^6$ ,  $4.0 \times 10^4$  and  $0.9 \times 10^1$  cfu  $g^{-1}$  for skin,  $3.2 \times 10^2$ ,  $1.0 \times 10^1$  and 0 cfu  $g^{-1}$  for muscle and  $1.0 \times 10^8$ ,  $2.2 \times 10^1$  and  $1.1 \times 10^1$  cfu  $g^{-1}$  for gills, respectively.

There were not significant differences between the results of the present study with the standard values ( $p > 0.05$ ) and this study shows that the overall microbial quality of fish encountered to the traditional way of transportation is found to be satisfactory.

The bacteriological status of the samples are presented in Table 1 and the comparison of the bacteriological status of the samples (cfu  $g^{-1}$ ) with the standard value is shown in Table 2.

This investigation presents the current status of microbial quality of south-harvested fish being sold in Kazeroon. From the total analyzed fish samples ( $n = 50$ ), 14.4% exceeded standard value of mesophilic aerobic

count, 4.0% crossed the standard value of total coliform count and 4.0% were beyond the standard value of *Vibrio parahaemolyticus* count. These results are generally similar to the findings of other researchers and can be supported by the research of Koutsoumanis *et al.* (2002) and Alam *et al.* (2006).

It seems that *Vibrio parahaemolyticus* which is a major cause of seafood-borne human disease occurred at very low frequencies in the samples of the present study. Although, *Vibrio parahaemolyticus* counts in these fish samples were not excessive, fish could pose a risk of *Vibrio parahaemolyticus* food poisoning if harvested in the summer period, when levels of *Vibrio parahaemolyticus* are highest in marine waters and it will be even worse if the summer-harvested fish is eaten raw or semi-raw.

This risk would be considerably reduced however, if fish are harvested during winter (Bouchriti *et al.*, 1994). *Vibrio parahaemolyticus* has been shown to be responsible for incidents of food poisoning when present in large numbers ( $10^6$  viable cells  $g^{-1}$ ) in seafood. Also, *V. parahaemolyticus* is a natural bacterial inhabitant of marine waters and hence its survival pattern in fish during storage is also of interest (Bouchriti *et al.*, 1994).

Vibrios are located in the sediments but they are resuspended when the temperature increases (Bryan, 1980; Kaneko and Colwell, 1973) in a previous study, only 11 (2%) of 582 isolates were identified as *V. parahaemolyticus*. The absence of *V. parahaemolyticus* could be attributed to its actual absence or to the relative selectivity of the TCBS agar used for its isolation (Balakrish *et al.*, 1980; Kourany, 1982).

Table 1: The bacteriological status of the fish samples (cfu  $g^{-1}$ )

Samples	No. of samples	Sampling area	Average		
			Total bacterial count (SV*: $10^6-10^7$ )	Coliform count (SV: $4.0 \times 10^2$ )	<i>Vibrio parahaemolyticus</i> count (SV: $10^2$ )
<i>Scorpaenopsis juttatus</i>	25	Skin	$5.5 \times 10^6$	$6.0 \times 10^1$	$0.9 \times 10^1$
		Muscle	$10^2$	$1.0 \times 10^1$	0
		Gills	$10^5$	$1.5 \times 10^1$	$1.2 \times 10^1$
<i>Otolithes ruber</i>	25	Skin	$10^5$	$2.0 \times 10^1$	0
		Muscle	$5.5 \times 10^2$	$1.0 \times 10^1$	0
		Gills	$10^8$	$3.0 \times 10^1$	$1.0 \times 10^1$

\*Standard value (cfu  $g^{-1}$ )

Table 2: Comparison of the bacteriological status of the fish samples (cfu  $g^{-1}$ ) with the standard value

Grade of samples	No. of samples	Sampling area	Samples exceeded standard value of TBC*		Samples exceeded standard value of Coliform		Samples exceeded standard value of <i>Vibrio</i>	
			Number	%	Number	%	Number	%
<i>Scorpaenopsis juttatus</i>	25	Skin	2	8	1	4	1	4
		Muscle	0	0	0	0	0	0
		Gills	4	16	1	4	0	0
<i>Otolithes ruber</i>	25	Skin	3	12	0	0	1	4
		Muscle	0	0	0	0	0	0
		Gills	2	8	1	4	1	4

\*TBC: Total Bacterial Count

Consequently, techniques for the isolation of *V. parahaemolyticus* from fish should be evaluated and improved, if necessary (Bouchriti *et al.*, 1994).

### CONCLUSION

Increase in the bacterial loads of some of these commercial fish may be attributed to the factors such as contaminated containers and mishandling. The multiplication of bacteria at ambient temperatures may also have contributed to this increase. Both fecal indicator bacteria and pathogens have been shown to multiply in marine products after harvest and prior to marketing (Cook and Rupie, 1989). But from the present study it is found that the changes in the level of bacterial counts from harvest to sale does not make the South-harvested fish to be exceeded the standard value. And it can be concluded that although, there are some defects in the traditional way of fish-transportation in Iran this way does not lead the product to reach the critical microbial point.

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