

OCCURRENCE OF *VIBRIO SPP.* IN BIVALVE MOLLUSCS HARVESTED FROM BUTRINTI LAGOON, ALBANIA

Sonila ÇOÇOLI^{1*}, Pranvera ÇABELI¹, Elvira BELI², Tana SHTYLLA¹

¹Agricultural University of Tirana, Faculty of Veterinary Medicine, Albania

²Agricultural University of Tirana, Faculty of Agriculture and Environment, Albania

*(Corresponding author: sonilacocoli@yahoo.com)

Abstract

The main production of molluscs in Albania consists in the cultivation of *Mytilus galloprovincialis*, which takes place in Butrinti lagoon. All the positive cases of *Vibrio* spp. were isolated between June-September, which is related to the fact that *Vibrio* species prefer high water temperatures. By analyzing the physical and chemical indicators of water it was observed that the above mentioned species of *Vibrio* were isolated in the average temperature of 26.2 °C, average pH of 8.42 and average salinity 28.97 ‰. Based on the results obtained from this study, *Vibrio* species isolated from Butrinti lagoon molluscs were adapted to the following physical-chemical water conditions: temperature intervals of 23.6-28 °C, pH of 8.15 to 8.8 and salinity of 22.1-33.7 ‰.

Key words: Bivalve molluscs, Butrinti lagoon, *Vibrio* spp.

Introduction

Fish and mussels result to be the second source of proteins for consumers behind meat products. However, all over the world have raised concerns about health risks derived from mussels contaminated with various pathogens (2). Although most reports of outbreaks of food infections came from the United States, there are several reports from Europe, Australia and Asia. Since the end of 1800 there has been over 400 epidemic diseases from food origin and over 14 000 cases of gastroenteritis associated with the consumption of contaminated shellfish (3).

Molluscs in Albania occupy a significant place among fishery products. The main production consists in the cultivation of *Mytilus galloprovincialis*, which takes place in Butrinti lagoon (4). This lagoon is located in southern Albania, with an area of 1600 ha and has a production capacity of about 4,500 tons of mussels / year (1).

Vibrios are microorganisms spread in coastal waters around the world and they can contaminate bivalve molluscs. *Vibrio* species are tolerant to salt so they can survive in marine waters and pose a serious threat to public health. They are pathogenic to humans and mortality by members of *Vibrionaceae* family is 10 times higher than that caused by *Salmonella* or *Escherichia coli* (8).

The aim of this study was to assess the presence and incidence of *Vibrio* spp. in bivalve mussels, as very important pathogens causing gastroenteritis in humans.

Materials and methods

This study was carried out on samples of bivalve mussels of species *Mytilus galloprovincialis* collected from Butrinti lagoon during the period January 2010 - December 2012, by analyzing a total of 234 samples. The samples were taken from 3 stations of the lagoon: North, South and West. All the analyses were performed in the laboratories of microbiological control for aquatic products at Food Safety and Veterinary Institute of Tirana.

Detection of *Vibrio spp.*

Detection of *Vibrio spp.* was performed according to ISO / TS 21872-1:2007 (7). For initial dilution the enrichment media Alkaline Saline Peptone Water (ASPW) was used. The representative sample (25 g) was homogenized with 225 ml ASPW. Further, this homogenate was incubated at 41.5 °C for 6 h ± 1 h. After 24 hours the culture obtained was inoculated with a loop to the surface of the plate with TCBS agar and *Vibrio* CROM agar, in order to allow the growth of well isolated colonies. The presence of opaque yellow colonies with a diameter of 2-3 mm in TCBS was indicative of the presence of *V. cholerae*, *V. alginolyticus*, *V. fluvialis* and/or, *V. metschnikovii*, while the growth of green colonies was indicative of the presence of *V. parahaemolyticus*, *V. vulnificus*, *V. harvey* or *V. mimicus*. In *Vibrio* CHROM agar the presence of smooth colonies with blue color indicated the presence of *V. cholerae* and smooth red colonies indicated the presence of *V. parahaemolyticus*. At the end of incubation, suspected colonies were subject to the following tests for confirmation: determination of oxidative activity, planting in tryptophan broth for indole reaction, halotolerance test (inoculation in Peptone Water with different salt concentrations), and API 20E.

Results and discussion

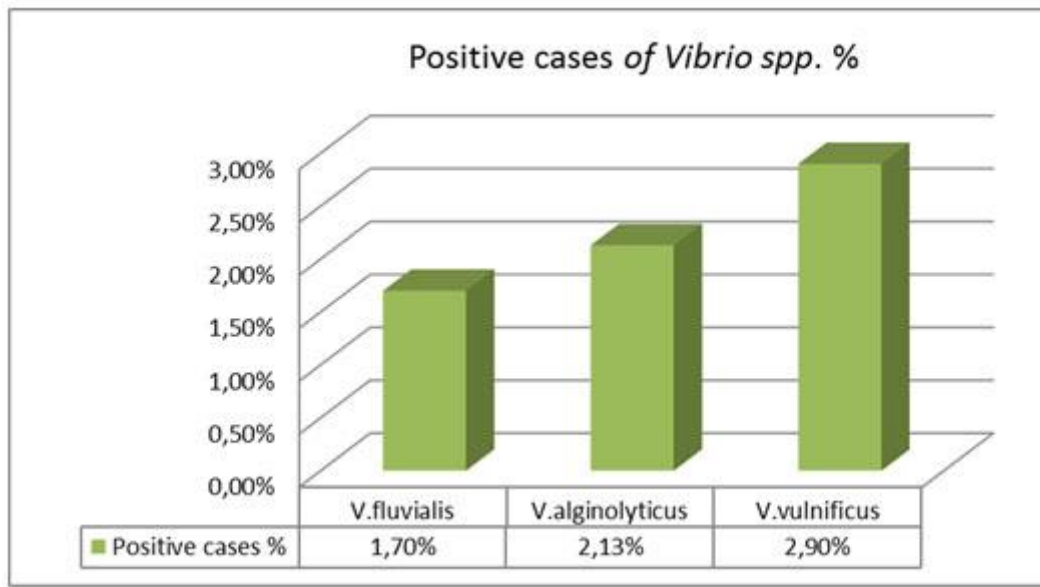
In this study, carried out during the period 2010-2012, the number and prevalence (%) of samples that resulted positive for the presence of *Vibrio spp.* were the following:

- 7 samples with *V. vulnificus* (2.9%);
- 4 samples with *V. fluvialis* (1.7%);
- 5 samples with *V. alginolyticus* (2.13%)

The table below shows the positive results of *Vibrio spp.* as well as the data on physic-chemical indicators of sea water, at the time of sampling.

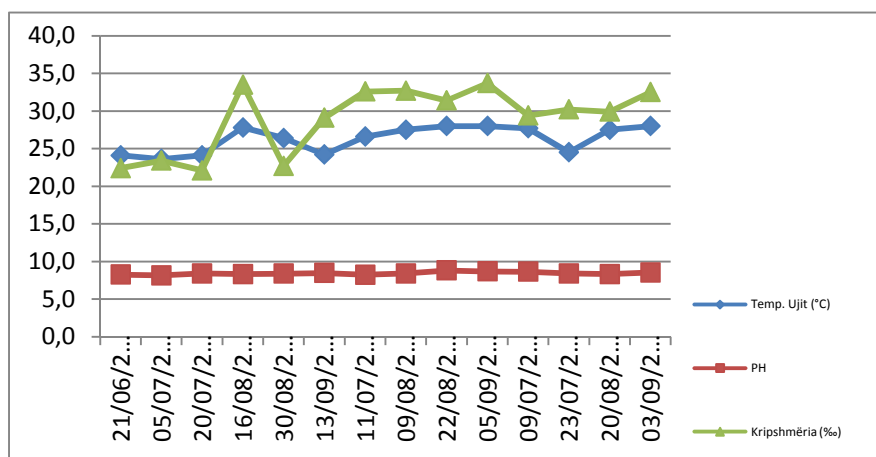
Date of sampling	<i>Vibrio spp.</i>			Water temp. (°C)	pH	Salinity (‰)
	<i>V. vulnificus</i>	<i>V. fluvialis</i>	<i>V. alginolyticus</i>			
21/06/2010		1	1	24.1	8.24	22.4
05/07/2010	1			23.6	8.15	23.4
20/07/2010	1			24.1	8.39	22.1
16/08/2010		1		27.8	8.32	33.5
30/08/2010		1	1	26.4	8.38	22.7
13/09/2010	1			24.2	8.47	29.1
11/07/2011		1		26.6	8.22	32.6
09/08/2011	1			27.5	8.4	32.7
22/08/2011	1			28	8.8	31.4
05/09/2011	1			28	8.67	33.7
09/07/2012			1	27.7	8.62	29.4
23/07/2012	1			24.5	8.4	30.2
20/08/2012			1	27.5	8.32	29.9
03/09/2012			1	28	8.52	32.5
TOTAL	7	4	5			

Percentage (on a total of 234 samples)	2.9 %	1.70 %	2.13 %			
Maximum				28.00	8.80	33.70
Average				26.2°C	8.42	28.97 ‰



As shown in the table and graph above, out of 234 samples analyzed in a three-year period. All the positive cases of *Vibrio* spp. were isolated between June-September.

By analyzing the physical and chemical indicators of water it was observed that the above mentioned species of *Vibrio* were isolated at the average values of 26.2 °C of temperature, 8.42 of pH and 28.97 ‰ of salinity.



Based on the results obtained from this study, *Vibrio* species isolated from Butrinti lagoon molluscs were adapted in physical-chemical conditions of water: temperature between 23.6-28° C, pH between 8.15 to 8.8 and salinity between 22.1-33.7 ‰.

Conclusions

Out of 234 samples analyzed those positive for the presence of *Vibrio* spp. were as follows: 7 samples positive for *V. vulnificus* (2.9%), 4 for *V. fluvialis* (1.7%), 5 for *V. alginolyticus* (2.13%). *V. vulnificus*, *V. alginolyticus* and *V. fluvialis* are among *Vibrio* species of interest for humans, as they can cause food poisoning (6).

Positive cases of *Vibrio* spp. were isolated during the period June-September, indicating that *Vibrio* species have a preference on high water temperatures (5).

Vibrio species isolated from Butrinti lagoon molluscs were adapted in physical-chemical conditions such as water temperature intervals 23.6-28° C, pH 8.15 to 8.8 and salinity 22.1-33.7 ‰.

References

- Anderson A. Bilodeau A. Gilg M. and Hilbish T. (2002). Routes of introduction of the Mediterranean mussel (*Mytilus galloprovincialis*) to Puget Sound and Hood Canal. *Journal of Shellfish Research*. **21** (1): 75-79.
- Arcangeli G. (2005). The problem of enteric viruses and vibrios. *Eurofishmarket* **1**: 4-10 (in Italian).
- Blake P., Weaver R., Hollins D. (1980). Diseases of humans (other than cholera) caused by vibrios. *Annual Review of Microbiology*. **34**: 341-367.
- Bego F. Memaj F. Peja N. Vangjeli J. Dharmo S. Ura A. Zotaj A. (2009). Management Plan of Butrinti Lagoon pp 51-53 (in Albanian).
- Corrain C. Arcangeli G. Fasolato L. Manfrin A. Rossetti E. Piazzzi E. Mioni R. Pavoni E. Lo-sio N. Sanavio G. Sufredini E. Croci L. (2007). Climatic and environmental influences on the presence of enteric viruses in bivalve molluscs. *Industria Alimentari*. **467**: 277-283 (in Italian).
- Desmarchelier P.M. (2000): *Vibrio*. In: *Encyclopedia of Food Microbiology*. Robinson R. Batt C. Patel P. pp 2237-2242.
- ISO/TS 21872-1. 2 (2007). Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of potentially enteropathogenic *Vibrio* spp. *Proceedings of the International for Standardization*. pp 5-22.
- Pruzzo C. Huq A. Colwell R. Donelli G (2005). Pathogenic *Vibrio* Species in the Marine and Estuarine Environment. In: *Oceans and Health: Pathogens in the Marine Environment: B Shimshon. R Colwell: 217-252.*