



# Vibrio parahaemolyticus

Family *Vibrionaceae*

Genus *Vibrio*

Bacterium

## Characteristics and sources of *Vibrio parahaemolyticus*

### Main microbiological characteristics

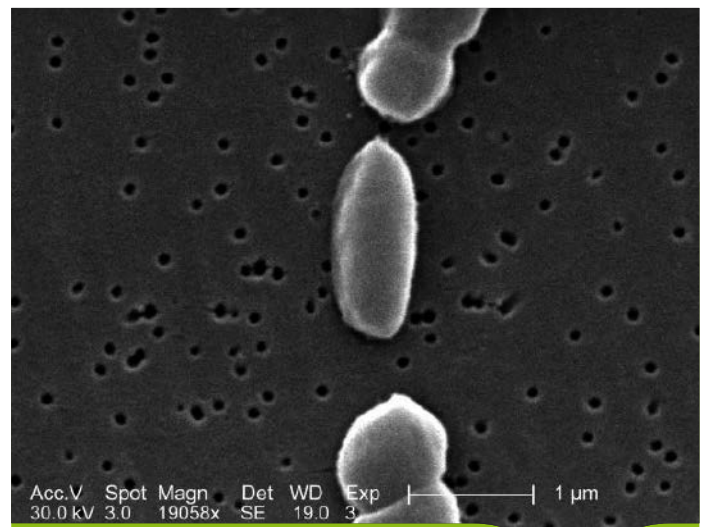
*V. parahaemolyticus* is a rod-shaped or curved Gram-negative bacteria, 0.5 to 1 µm in diameter, halophilic (grows in 0.5 to 10% NaCl), oxydase positive, sucrose negative and facultatively anaerobic. Foodborne *V. parahaemolyticus* infections are mostly present in the form of gastroenteritis.

The conditions influencing the growth of this species of bacteria are given in the [Table 1](#).

**Table 1.** Characteristics for survival/growth of *Vibrio parahaemolyticus*

Parameters	Optimum	Extremes
Temperature (°C)	37	5-43
pH	7.8-8.6	4.8-11
Water activity (a <sub>w</sub> )	0.981	0.940-0.996
NaCl (%)	1.5-3	0.5-10

It has been shown that the pathogenicity of enteropathogenic strains is related to the presence of at least one of the two following haemolysins, TDH (Thermostable Direct Haemolysin) and TRH (TDH-Related Haemolysin), produced in the human digestive tract. They have similar lytic, cytotoxic and enterotoxic activity. In the natural environment, strains carrying haemolysin genes are rare and usually account for only 0.2 to 2% of *V. parahaemolyticus* strains isolated, although this figure can reach 15% in certain particular ecosystems. On the other hand, genes coding for at least one of the two haemolysins are found in 95% of strains of *V. parahaemolyticus* isolated in the stools of patients suffering from gastroenteritis. To date, 13 O antigens and 71 K antigens have been identified in strains of clinical origin. A pandemic clone of *V. parahaemolyticus*, belonging to the serotype O3:K6, appeared in Bangladesh in 1996 and spread rapidly in India, South-East Asia, Japan, North America and Europe. New pandemic clones derived from this O3:K6 clone have been identified since 1996.



*Vibrio parahaemolyticus* © CDC/ Janice Carr

### Sources of the hazard

The natural habitat of *V. parahaemolyticus* is in estuaries and coastal waters throughout the world. This bacterial species is often found in sediments, plankton, fish, crustaceans and bivalve molluscs, especially oysters and mussels. Water temperature and salinity play an important role in the growth of *V. parahaemolyticus*. The highest densities of this bacterium are encountered in water temperatures above 18-20°C and in intermediate salinities (i.e. between freshwater and saltwater). Peaks of *Vibrio* counts are observed during the hottest months. In the cool season, some vibrios subsist in sediments and plankton as "viable non-culturable" forms.

### Transmission routes

*V. parahaemolyticus* food infections are mainly caused by consuming raw or undercooked fish or shellfish, or by cross-contamination after cooking. Exceptional cases of extra-intestinal infections by the dermal route have been reported, following contamination of open wounds by seawater, or injuries while handling seafood.

### Recommendations for primary production

- Observe good hygiene practice.
- Ensure that shellfish spend as little time as possible out of water at ambient temperatures.
- Fully comply with regulatory temperatures during handling and transport.

## Foodborne human illness

### Nature of the disease (Table 2)

The vast majority of *V. parahaemolyticus* infections manifest by gastroenteritis. The TDH and/or TRH haemolysins may contribute to the poisoning but only if they are produced in the human digestive tract.

**Susceptible population group(s)**<sup>(1)</sup>: Clinical manifestations can be severe (dysentery-like syndrome, septicaemia) in very young children, the elderly, and subjects with an underlying disease (especially liver disease) or with a suppressed immune system.

### Dose-effect<sup>(2)</sup> and dose-response<sup>(3)</sup> relationships

A dose-response relationship established by the FDA indicates that the dose of *V. parahaemolyticus* pathogens causing illness in 1% of people exposed is in the order of 10<sup>5</sup> bacterial cells.

## Epidemiology

### Surveillance systems

The number of cases observed can vary significantly, depending on the surveillance system set up in each country. For example, several cases are reported every year in the United States, where non-cholera *Vibrio* infections are notifiable illnesses. In France, as in the other European countries, it is difficult to know their incidence, especially for the less severe forms, as these infections are not notifiable and are also probably under-diagnosed. As a result, their frequency is likely to be underestimated.

### Epidemiological data

Outbreaks have been reported, mainly during the hottest months, in South-East Asia and North America. *V. parahaemolyticus* represents a serious public health problem in these countries because of the high consumption of seafood, especially raw, and is recognised to be the principal cause of gastroenteritis associated with the consumption of seafood.

Inversely, *V. parahaemolyticus* infections are reported less frequently in Europe:

- a few sporadic cases have been recorded in Spain, where an outbreak (64 patients) related to the consumption of oysters occurred in 1999; more recently, in July 2004, an outbreak was reported associated with the consumption of boiled crab involving 80 people;
- in France, *V. parahaemolyticus* was detected in the Arcachon Basin in 1988. An epidemic of *V. parahaemolyticus* was recorded in 1997 in an army regiment in the Var département, probably related to the consumption of mussels or shrimps in a sauce. In 2001, an outbreak (100 cases) was linked to the consumption of mussels from Ireland. In 2009, four people were affected by an outbreak linked to the consumption of mussels, and the strains isolated carried the gene coding for the TDH haemolysin and belonged to the O3:K6 serogroup.

Between 1995 and 2012, the National Reference Centre for *Vibrio* and cholera at the Institut Pasteur recorded 20 cases of gastroenteritis, 3 septic shocks and 2 superinfections of a wound, due to this bacterium (Table 3). All the strains responsible for gastroenteritis carried the gene coding for either the TDH or TRH haemolysin, one strain carried both. Seven cases of gastroenteritis were associated with the O3:K6 clone.

(1) Susceptible population group: people with a higher than average probability of developing symptoms of the disease, or severe forms of the disease, after exposure to a foodborne hazard [definition used in ANSES data sheets].

(2) Relationship between the dose (the quantity of microbial cells ingested during a meal) and the effect on an individual.

(3) For a given effect, the relationship between the dose and the response, i.e. the probability of this effect appearing in the population

Table 2. Characteristics of the illness

Mean incubation period	Target population	Main symptoms	Duration of symptoms	Duration of contagious period (excretion)	Complications	Asymptomatic forms
12 – 24 hours (may vary between 4 and 96 hours)	Anyone consuming raw or insufficiently cooked seafood is potentially exposed.	Abdominal pains, cramps, watery diarrhoea, in some cases bloody diarrhoea. Nausea, vomiting and fever may also occur. The illness is frequently benign or moderate, although some cases have required hospitalisation.	1 to 3 days on average, but can extend to 7 days	Not applicable	Some cases require hospitalisation. Exceptionally, <i>V. parahaemolyticus</i> causes septicaemia in immune-suppressed subjects or those suffering from underlying diseases.	Because of the absence of systematic screening or investigation, there is no information available concerning asymptomatic human carriers.

Table 3. Strains of non-cholera *Vibrio* responsible for human infections in mainland France. National Reference Centre (NRC) 1995-2012

Species	Number of strains received at the NRC	Clinical forms (number of cases)	Number of fatalities	Context of contamination
<i>Vibrio parahaemolyticus</i>	25	Gastroenteritis (20)	-	Consumption of seafood (13), ND (6), NE (1)
		Septicaemia (2)	1	ND (2)
		Various forms of suppuration (2)	-	Consumption of seafood (1), contact with seafood (1)
		Wound + septicaemia (1)	1	Contact with seafood (1)

ND: non-documented, NE: context of the contamination not established. These data were obtained as part of the activities of the National Reference Centre for *Vibrio* and cholera, commissioned by the Ministry of Health and funded by the French Institute for Public Health Surveillance and the Institut Pasteur. All rights reserved.

# Role of food

## Main foods to consider

### Conditions leading to the contamination of produce

The sources of contamination are: the natural environment, cross-contamination during handling, contamination by washing with contaminated seawater, and recontamination after cooking. Any break in the cold chain can contribute to the development of *V. parahaemolyticus* present in produce and therefore to an increase in the contamination level. The time taken to process produce and the temperature of the premises play an important role in the development of this bacterium: when exposed for 2-3h at ambient temperature, *V. parahaemolyticus* can be observed to grow at  $10^2$ - $10^3$  cfu/g and even above  $10^5$  cfu/g.

### Foods implicated

Most cases of gastroenteritis are related to the consumption of seafood:

- shellfish: oysters, mussels,
- crustaceans: shrimps, crabs,
- raw fish.

## Inactivation treatments in industrial environments

Different methods have been shown to be effective and can be applied in industry (Table 4).

Table 4. Inactivation treatments in industrial environments

Disinfectants	Thermal treatments
<i>Vibrio</i> are sensitive to many disinfectants authorised in the agro-food industry, on condition users follow the instructions for use.	<b>Heat</b> These bacteria are extremely heat-sensitive and are destroyed rapidly in temperatures higher than the maximum temperature for growth; the bacterium can therefore be inactivated effectively by cooking the produce sufficiently. However, the time necessary for total inactivation by cooking depends on the size of the initial bacterial population. At 52°C for 7.8 min, a 5 log <sub>10</sub> reduction is observed in oyster flesh. <b>Cold</b> Cold storage limits growth but does not eliminate <i>V. parahaemolyticus</i> . After 1 month at 20°C, a single log <sub>10</sub> reduction is observed in oyster flesh.
High Pressures	Ionisation
High pressure treatment destroys the bacteria without altering the nature of the food. Oyster flesh subjected to 345 MPa for 2 minutes undergoes a 5 log <sub>10</sub> reduction.	Ionisation treatment (irradiation) has been shown to be effective in reducing the number of bacteria. At 1 kGy, a 6 log <sub>10</sub> reduction is observed in oyster flesh.

## Monitoring in food

### Regulations in force

European legislation (Regulation (EC) 2073/2005 and the acts amending it) does not currently propose specific microbiological criteria in food regarding human pathogenic *Vibrio*, but recommends that reliable methods be developed for assessing the risk related to *Vibrio parahaemolyticus* in seafood. In France, however, the Ministry for Agriculture and Fisheries can demand that seafood to be imported be screened for *Vibrio* pathogenic to humans. A memorandum (DGAL SSA373/72 of October 2004) addressed to the veterinary services, especially those responsible for border inspection stations, lays down that "batches of imported fishing produce found to be contaminated by a strain of *Vibrio parahaemolyticus* carrying haemolysin genes (TDH and/or TRH) must be withdrawn and destroyed in compliance with Regulation (EC) 1774/2002". Only the quality aspect is considered.

It should be noted, however, that no systematic surveillance or inspection plan is specified by the Directorate General for Food (DGAL) or the Directorate General for Competition Policy, Consumer Affairs and Fraud Control (DGCCRF) for bacteria of the genus *Vibrio*.

## Methods for detection and counting

There is currently no standard reference method available for counting *Vibrio* in seafood.

In France, analytical laboratories generally use either a "provisional protocol" drafted jointly by the ANSES laboratory at Boulogne-sur-Mer, the NRC for *Vibrio* and Cholera, and the EHESP at Rennes, which is particularly suitable for the analysis of crustaceans and fresh fish (but which has not been validated for live shellfish) or the technical specification ISO/TS 21872 for the detection of potentially enteropathogenic *Vibrio* spp.. The ISO technical specification is currently being reviewed. The intention is to give a broader role to molecular biology techniques in this standard.

Methods involving conventional or real-time PCR for the detection, quantification and characterisation of *V. parahaemolyticus* have been developed or are under development. These methods are based on screening for genes characteristic of the species *V. parahaemolyticus* (especially the *toxR* gene or the R72H sequence) and genes coding for the TDH and TRH haemolysins.

### Recommendations to operators

- Minimise the time between removing oysters from water and refrigeration.
- Fully comply with good hygiene practice. In applying HACCP principles, consider the hazard regarding *Vibrio*, including studies of the prevalence of the hazard.
- Fully comply with regulatory temperatures during handling and transport, and also for presentation at points of sale. Recommendations for shellfish include maintaining them at temperatures below 10°C.

## Domestic hygiene

### Recommendations to consumers

- Bear in mind that the consumption of seafood in summer increases the risk of *Vibrio*-related gastroenteritis.
- In summer, transport shellfish and seafood in isothermal containers (iceboxes) and transfer rapidly to the refrigerator (4°C).
- Respect good hygiene practice when handling and preparing foods:
  - consume within two hours of removing them from the refrigerator;
  - avoid contact between cooked and raw seafood to limit cross-contamination.
- Patients suffering from underlying diseases, chronic liver conditions (hepatitis, cirrhosis, alcoholism) or diseases resulting in iron overload, and immunosuppressed patients (diabetes, cancer) with increased susceptibility to infection by *Vibrio*, should avoid consuming raw or insufficiently cooked seafood (e.g. oysters, mussels, clams or shrimps).

# References and links

## General references

- ANSES (2012). Avis et rapport de l'Anses relatifs à une demande d'évaluation du risque lié à *Vibrio parahaemolyticus* via la consommation de produits de la mer. [Risk assessment of *Vibrio parahaemolyticus* in seafood]  
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- FAO/WHO [Food and Agriculture Organization of the United Nations/World Health Organization]. 2002. Risk assessment of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood. Report of a Joint FAO/WHO Expert Consultation (Bangkok, Thailand). 59p.  
<http://www.who.int/foodsafety/publications/micro/aug2002.pdf>
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- Quilici ML, Robert-Pillot A (2011). Infections à vibrions non cholériques. EMC (Elsevier Masson SAS, Paris), Maladies infectieuses 8-026-F-15.
- Thompson, F.L., Austin, B., Swings, J. (eds). 2006. Biology of Vibrios. ASM Press, Washington, DC.

## Useful links

- Public Health Agency of Canada, pathogen safety data sheet:  
<http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/vibrio-parahaemolyticus-eng.php>
- Centers for Disease Control and Prevention, general information on bacteria:  
<http://www.cdc.gov/nczved/divisions/dfbmd/diseases/vibriop/>
- New Zealand Food Safety Authority, datasheet on *Vibrio parahaemolyticus*:  
[http://www.foodsafety.govt.nz/elibrary/industry/Vibrio\\_Parahaemolyticus-Science\\_Research.pdf](http://www.foodsafety.govt.nz/elibrary/industry/Vibrio_Parahaemolyticus-Science_Research.pdf)
- US Food and Drug Administration, "Bad Bug Book":  
<http://www.fda.gov/Food/FoodSafety/Foodbornellness/FoodbornellnessFoodbornePathogensNaturalToxins/BadBugBook/ucm070452.htm>
- Soochow University (Taiwan), datasheet on *Vibrio parahaemolyticus*:  
<http://www.scu.edu.tw/microbio/vp-eng.htm>

## Reference laboratories (NRC, EURL, NRL)

- National Reference Centre for *Vibrio* and Cholera: Institut Pasteur, Paris.
- National Reference Laboratory for *Vibrio* spp. in fishery products: ANSES, Boulogne-sur-Mer Laboratory for fishery products.
- National Reference Laboratory for shellfish microbiology: French Research Institute for Exploitation of the Sea (IFREMER) – Nantes.
- European Union Reference Laboratory for monitoring the viral and bacteriological contamination of bivalve molluscs: The laboratory of the Centre for Environment, Fisheries and Aquaculture Science (Cefas) Weymouth, United Kingdom.