

# A temperature dependent disease : impact of *Vibrio harveyi* on the abalone *Haliotis tuberculata*

Agnès Travers<sup>1,2\*</sup>, O. Basuyaux<sup>2</sup>, M. Garnier<sup>3</sup>, J.L. Nicolas<sup>3</sup>, M. Koken<sup>1</sup>, S. Huchette<sup>4</sup> and C. Paillard<sup>1</sup>

<sup>1</sup> IUEM, Plouzané, France, <sup>2</sup> SMEL, Blainville sur mer, <sup>3</sup> Ifremer Brest  
<sup>3</sup> France Haliotis, Plouguerneau, France. \* marie-agnes.travers@univ-brest.fr

## Introduction:

Wild abalone stocks are declining worldwide due to over-exploitation or diseases affecting several *Haliotis* species, including the ormer *Haliotis tuberculata*, the most commercially-important abalone in Europe [1].

*Vibrio carchariae* has been isolated in *H. tuberculata* during the mass mortality that occurred in 1998, in France [2]. Mortalities seems to be directly link to seawater temperature (>16,5°C, Figure 1). This vibrio is a synonym of *Vibrio harveyi*, already known to be pathogenic for shark, shrimp and prawn [3].

The goal of this new research topic is to highlight direct implications of this bacterium in ormer mortalities and to elucidate mechanisms of interaction between *Vibrio harveyi* and *Haliotis tuberculata*.

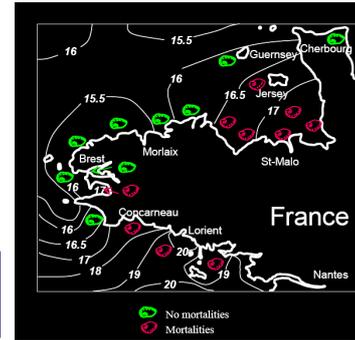


Figure 1: Distribution of *H. tuberculata* summer mortalities along France coast and isotherm of summer temperature maximum. Red abalone correspond to witnessed mortalities

Huchette and Clavier [1]

## 1. Pathogenesis: Clinic signs

Mortalities have been reported in the field and in hatcheries in Brittany and Normandy (France). Mortalities are associated with loss of muscular motility, pericardial inflammation and white pustules on the foot (corresponding to bacteria clumps, as shown by histology, bacterial isolation and PCR identification).

Bacteria were principally found in muscle and haemolymph.

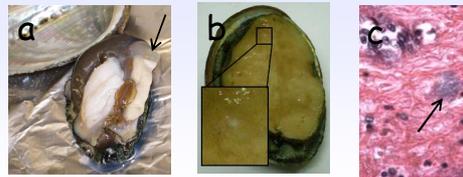


Figure 2: Macroscopic and microscopic characteristics of mortalities associated with *V. harveyi*. a) Pericardial inflammation, b) White pustule, c) Hematoxylin Eosin coloration of muscle section.

## 2. Modulation of the interaction

### 1. Pathogenicity of different *V. harveyi* strains

Different *V. harveyi* (isolated in field as in hatcheries during mass mortalities) were compared with collection strains. Their pathogenicity was tested *in vivo*.

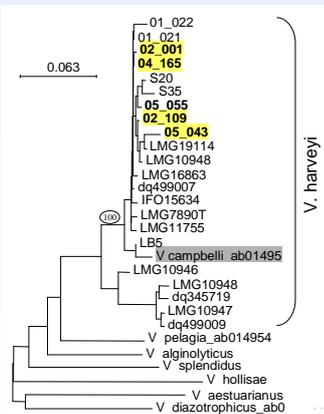


Figure 3: gyrB. Pathogenic *V. harveyi* are in yellow. 29 species, 808 sites (global gap removal), Neighbor Joining Method, Jukes and Cantor distance, 500 bootstrap replicates.

01\_021 to 05\_043 correspond to abalone isolated *V. harveyi*.

→ Pathogenic strains cannot be differentiated from non pathogenic ones on the basis of gyrB or rpoA sequences.

### 2. Pathogenicity *in vivo* : Temperature effect

As temperature seems to be determinant for disease development in the field, experimental inoculations were performed at different temperatures on hatchery abalone.

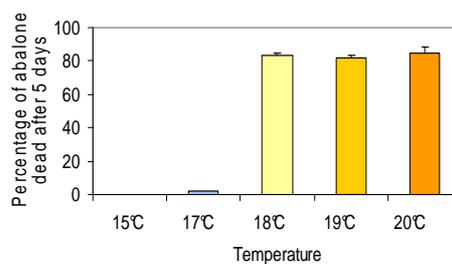


Figure 4: Percentages of dead abalone 5 days after introduction of *V. harveyi* strain 02\_001 into the surrounding water. (10<sup>6</sup> bact./ml during 24 hours, triplicate tanks, 20 abalone/tank).

→ Significant mortalities due to *V. harveyi* occurred only over 18°C.

### 3. Host physiology and maturation effects

As mortalities occur in the field simultaneously with spawning, effects of gonad maturation on susceptibility to *V. harveyi* was investigated on adult abalone at 19°C.

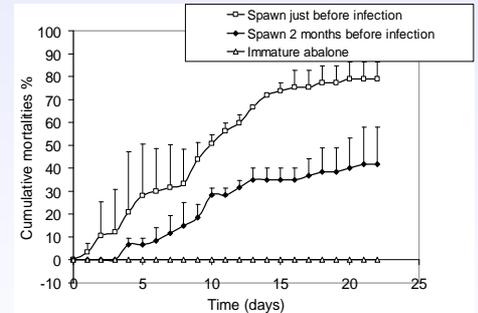


Figure 5: Evolution of mortalities after introduction of the pathogen *V. harveyi* strain 02\_001 into the water (10<sup>6</sup> bact./ml during 24 hours at 19°C, triplicate tanks, 20 abalone/tank).

→ Mature abalone (spawning immediately before infection) are more sensitive than abalone that spawned 2 months before. Immature abalone, independently of their size and age, appeared insensitive to infection.

## Conclusion and perspectives:

*H. tuberculata* mortalities associated with *V. harveyi* depends on *V. harveyi* strain, abalone age, gonad maturation level and water temperature. Abalone are affected by temperature for sexual maturation or growth. Temperature may also increase their sensitivity to bacteria. But it may also affect bacterial growth (optimum 28°C) and virulence.

Results highlight the importance of 1°C variation on disease development (17°C vs 18°C). Thus, in view of the current global warming, the impact of temperature on *V. harveyi* growth and virulence, and on abalone immunity should be investigated further. Moreover, differentiating virulent and avirulent strains should be one of our priority.

### Useful references :

- Huchette, S. M. H. and J. Clavier (2004). "Status of the ormer (*Haliotis tuberculata* L.) industry in Europe." *Journal of Shellfish Research*, 23(4) 951-5.
- Nicolas, J. L. et al. (2002). "*Vibrio carchariae*, a pathogen of the abalone *Haliotis tuberculata*." *Diseases of Aquatic Organisms* 50(1): 35-43.
- Austin, B. and Zhang, X.H. (2006) "*Vibrio harveyi* a significant pathogen of marine vertebrates and invertebrates." *Letters in applied microbiology* 43: 119-24.