MARINE MOLLUSC PATHOGENS

LABORATORY DIAGNOSTIC METHODS

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GENERAL FACT SHEET **B**

Laboratory techniques make it possible to **detect** pathogens present in marine molluscs: **histology ❶, bacterial cultures ❷ and molecular biology techniques**, in particular, **the polymerase chain reaction ❸.**

These diagnostic methods are called “direct” because they target the pathogen itself. PCR methods target the genetic material (DNA or RNA) of pathogens.

Once detected, the pathogen can be more precisely characterised using two other molecular biology techniques, frequently employed for determining a species or strain:

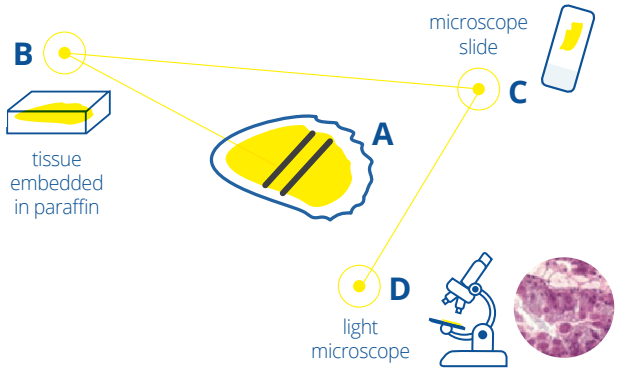
- **sequencing** gives the exact composition of the amplified pathogen DNA or RNA fragment;
- ***in situ* hybridisation** helps localise the target DNA or RNA (of the pathogen) in a tissue section observed under a microscope.

Mollusc mortality events are often linked to a combination of several factors.
The presence of a pathogen is not necessarily the sole cause of mortality.
A pathogen can be detected in molluscs outside of mortality events.

1 HISTOLOGY OR THE STUDY OF BIOLOGICAL TISSUES

(OBSERVED UNDER A MICROSCOPE)

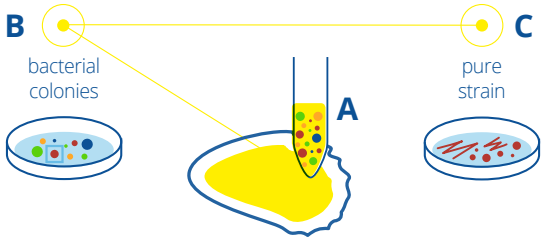
This method allows detecting the presence of certain pathogens as well as the lesions that they cause in marine mollusc tissues.



A piece of mollusc tissue **(A)** *is placed in a chemical fixative and then embedded in paraffin* **(B)** *to be able to cut it into thin sections. These sections are mounted on a slide* **(C)**, *stained and then observed under a microscope* **(D)**.

2 BACTERIAL CULTURES

This method makes it possible to culture and isolate predominant bacteria present in marine molluscs and in seawater, for subsequent identification.

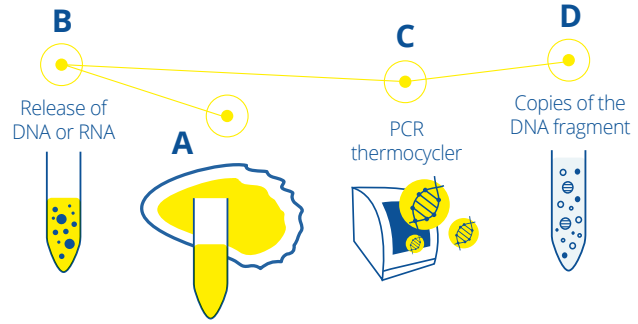


Mollusc tissues are ground **(A)**. *The ground tissue is spread on an agar plate containing all the nutrients needed for bacterial growth. The bacteria reproduce and form colonies* **(B)**. *Each single colony is spread on a new agar plate to obtain pure strains* **(C)**.

Pure strains can then be analysed using PCR, see method ❸.

3 POLYMERASE CHAIN REACTION (PCR)

This molecular biology technique is used to identify pathogens. It can amplify a targeted DNA or RNA fragment millions of times to obtain a sufficient quantity for detection.



Mollusc tissues are sampled and placed in a tube **(A)**. *Reagents are added to ‘digest’ the tissues and release the nucleic acids (DNA or RNA) contained in the cells* **(B)**. *PCR analysis consists of a succession of reactions to multiply the DNA fragment of interest* **(C)** *so that it can be detected* **(D)**.