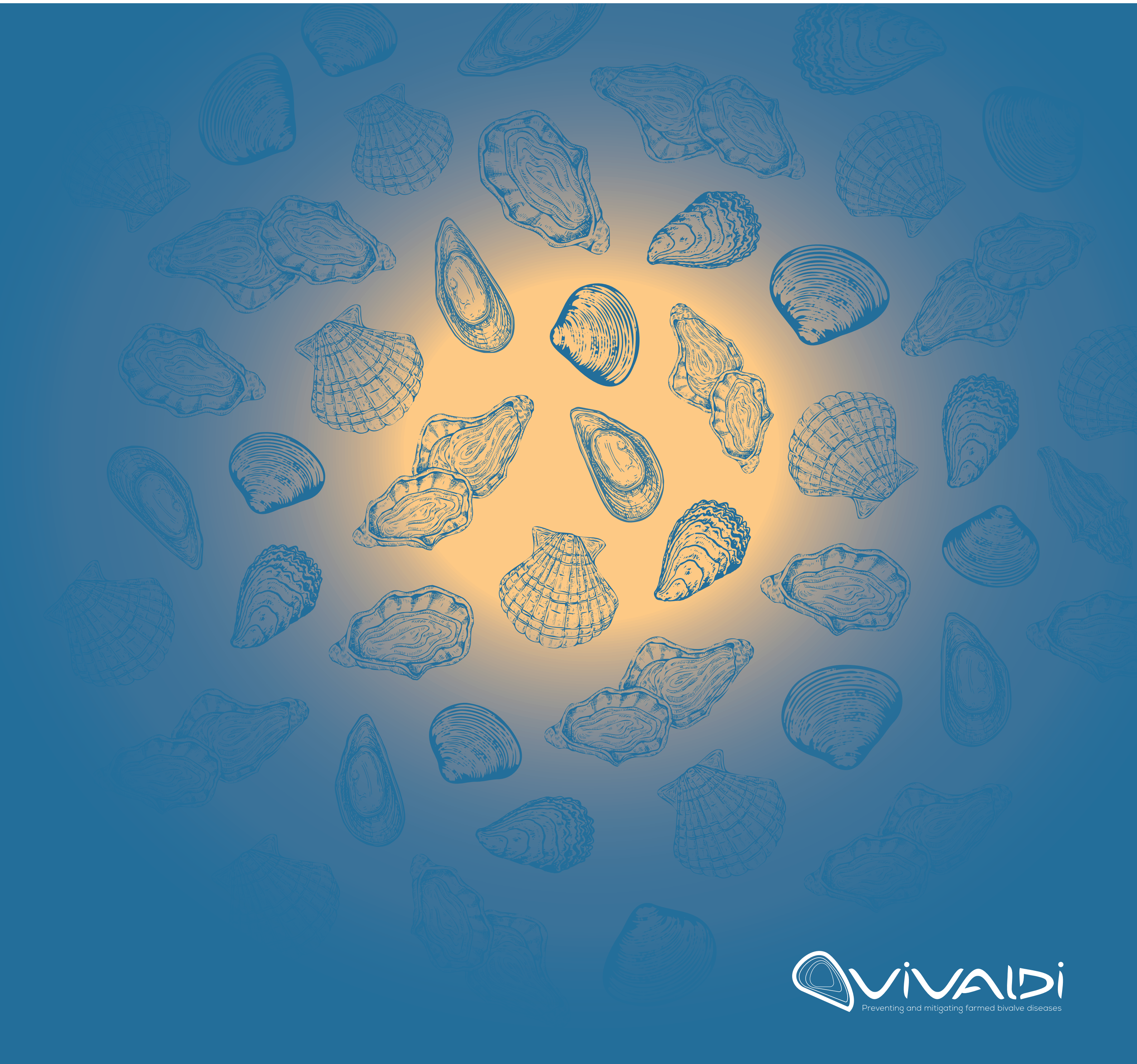
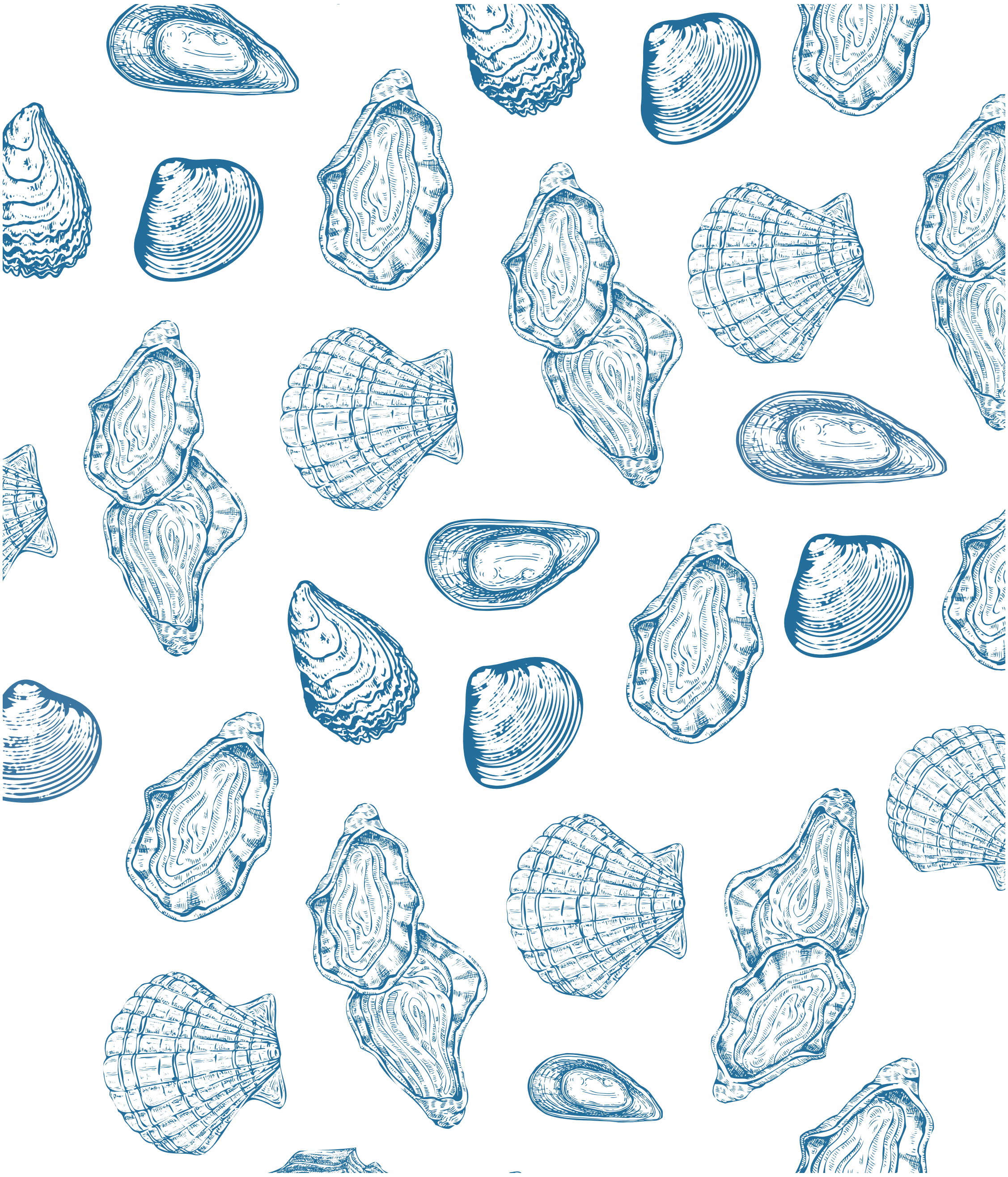
MANUAL

for bivalve disease management and biosecurity



VIVALDI
Preventing and mitigating farmed bivalve diseases



MANUAL FOR BIVALVE DISEASE MANAGEMENT AND BIOSECURITY

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HOW TO CITE:
Arzul I, Furones D, Cheslett D, Gennari L, Delangle E, Enez F, Lupo C, Mortensen S, Pernet F and Peeler E (2021)
Manual for bivalve disease management and biosecurity - H2020 VIVALDI Project - 44p

LEAD PARTNER FOR OUTPUT:
Ifremer and IRTA

ACKNOWLEDGEMENT:
The work described in this project has been funded by European Commission under the Horizon 2020 Framework Programme (www.vivaldi-project.eu/).

Design and print by: nyl communication
1ST EDITION: 2021

Oyster farms, Dungarvan Bay, Ireland.
Picture: ©VIVALDI

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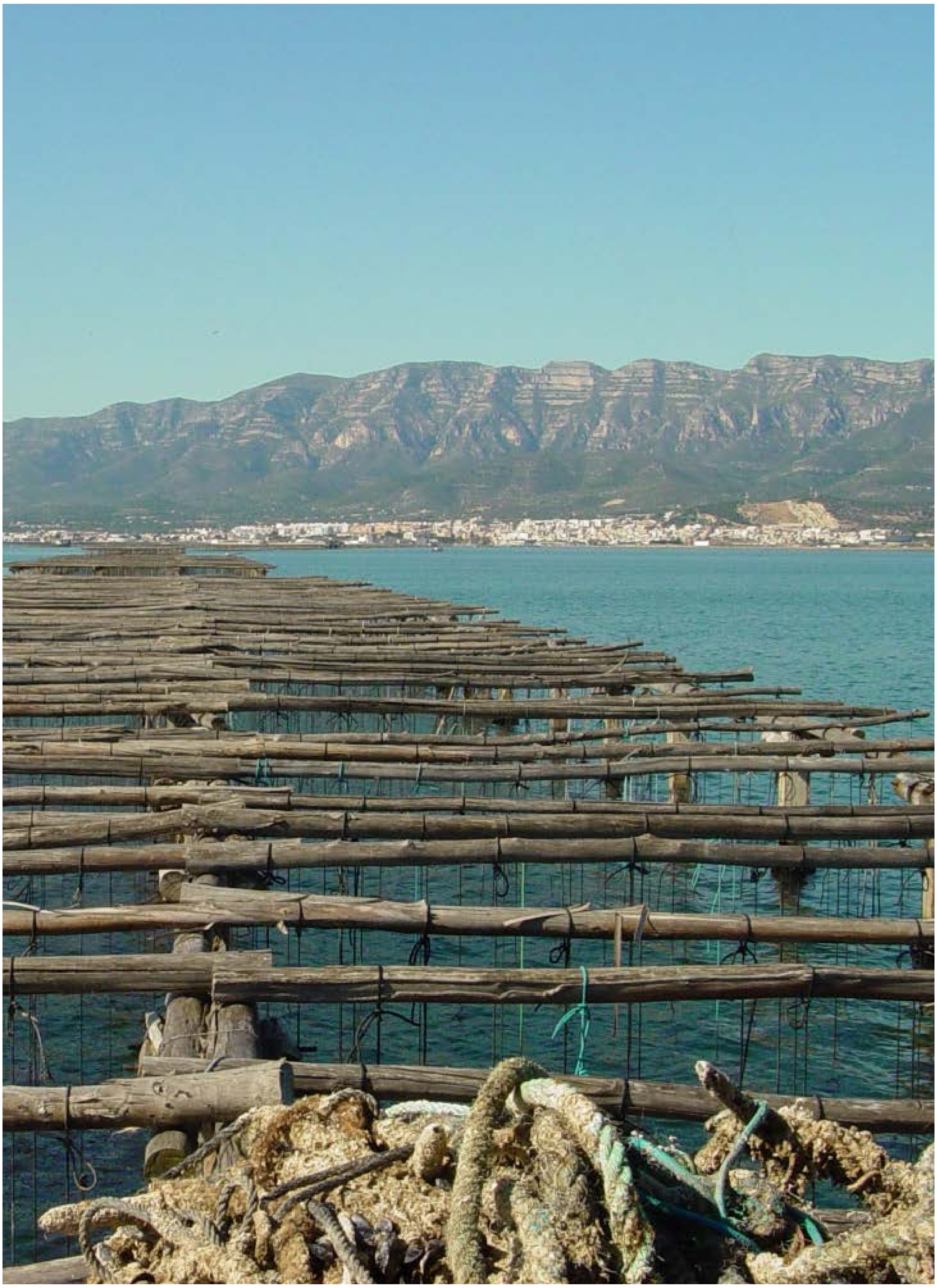

Mussel rafts Alfacs Bay, Delta del Ebro, Spain.
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Stakeholders’ meeting in Paris, 07 March 2019: preparing the ground for a biosecurity manual.
Picture: ©VIVALDI

INTRODUCTION

Shellfish farming is a vital economic sector in Europe, employing more than 40,000 people. However, it must cope with recurring episodes of mortality. For instance, the OsHV-1 virus has been responsible for high rates of mortality in juvenile cupped oysters in various European Union member states, especially since 2008. Another pathogen, the bacteria *Vibrio aestuarianus*, has been linked to mortality episodes affecting adult cupped oysters in France and Ireland. Other farmed mollusc species have not been spared: for instance, the cockle populations in Galicia which have dramatically declined, linked to the presence of a parasite called *Marteilia cochillia*.

Between 2016 and 2020, the VIVALDI project (https://www.vivaldi-project.eu/) aimed to improve the sustainability and competitiveness of the European shellfish industry, which was hit by a growing number of mortality cases over the recent years. To this end, tools and strategies to better prevent and mitigate the impact of bivalve diseases have been developed. For example, environmental approaches such as passive sensors or magnetic beads and electrochemical biosensors could be useful for pathogen surveillance and the development of early warning systems. Stimulating bivalve immunity has been shown to be possible and could be of interest for hatcheries-nurseries. Work has been achieved to optimize breeding programs for oysters as well as clams. The impact of environmental parameters on the development of bivalve diseases has been studied, allowing researchers to identify conditions favoring or mitigating disease development. Best husbandry practices to reduce mortality have been identified from the literature and field studies. UV treatments were successfully used to remove pathogens, oyster gametes and larvae from the wastewater. A risk ranking shellfish farm model was designed and is now ready to be used by the competent authorities to implement risk-based surveillance of shellfish diseases.

Most of these results have led the consortium to identify recommendations to better prevent, mitigate and control bivalve diseases.

A co-construction process involving scientists, decision-makers, hatcheries and producers from the main European producing countries was considered as the best approach in order to make this manual relevant and easy-to-use for the greatest possible number of stakeholders. The biosecurity manual is aimed to have a long-term impact on the end-users' practices and biosecurity in shellfish farming. It does not have regulatory goals but it aims to provide technical advices to assist implementing the legislation. When covering farming activities, recommendations identify best practices that need to be adjusted taking into account geographic and species specificities.

For each recommendation, a brief description is provided as well as the benefits and main limitations. Recommendations are organized by section and, then, by category of actions. Three main sections have been identified: communication issues, governance issues and technical issues. In addition, a glossary has been compiled to provide the precise definitions of the terms used.

Sample of clams, Chiogga, Italy.
Picture: ©VIVALDI

COMMUNICATION ISSUES

Stakeholders' meeting in Paris, 07 March 2019: preparing the biosecurity manual.
Picture : ©VIVALDI

Training, knowledge transfer, information and methodology

Examination of oyster cells using an epifluorescence microscope.
Picture: ©VIVALDI

1. TECHNOLOGY TRANSFER, TRAINING AND EXCHANGE OF BEST PRACTICES ON DISEASE RISK-MANAGEMENT

This is primarily the responsibility of the competent authority, working in collaboration with the industry and training institutions.

DESCRIPTION :

- **Working groups** or organizations, representing all stakeholders including training bodies at different scales (local, regional, national, EU and international) should be established. Education/training organisations must be included in these working groups.

    Stakeholders' organigrams including roles and contacts should be regularly updated.
    A "common language" is needed. All parties must make an effort to be understood by the other parties.
    Coordinators /moderators could contribute to facilitate information exchanges, e.g. defining technical words and concepts.

- Different tools could be used to improve communication between stakeholders:

    The development of an App to assist fluid and interactive communication system;
    The implementation of calendars and agendas to ensure regular contact and sustainability of working groups.

- Different supports can be used to share information about best practices and biosecurity:

    A manual distributed to stakeholders, in their mother tongue;
    Online training courses;
    A model for training courses;
    Single point of access (one-stop shop) for stakeholders to quickly locate relevant information and to request further information or training courses.

- **Training programmes** dedicated to producers, competent authorities, diagnostic laboratories should be defined collectively with all the stakeholders including education/training organizations.

BENEFITS :

- Increased knowledge among stakeholders will result in better biosecurity and a more sustainable production.
- Understanding each other's role and responsibilities will facilitate stakeholders' commitment to disease prevention and mitigation.
- Implementing the plans for fighting diseases, in a quicker and more efficient way.
- A more sustainable production will lead to a better productive output.

THE MAIN LIMITATIONS :

- There is no established model for training.
- The different roles and responsibilities of stakeholders for training, knowledge exchange etc. need to be agreed.
- Economic cost of training.
- Need to improve the information flow.

2. INFORMING STAKEHOLDERS ABOUT DISEASE STATUS AND RISKS

This is primarily the responsibility of the competent authority, working in collaboration with the shellfish industry.

DESCRIPTION :

- Establishment of communication channels between producers and Competent Authorities to allow for effective sharing and dissemination of information on shellfish health status and potential emerging pathogens* risks to all stakeholders. This relates to both notifiable and non-notifiable pathogens/diseases.

- Information which should be shared includes:

    Details of regulations and requirements under which shellfish producers must operate including changes to legislation, requirements in terms of notification;

    All available information on the **epidemiological status** (presence/absence, prevalence of a pathogen) of production/harvesting areas and those of potential suppliers (imports);

    Up to date **epidemiological maps** (distribution and prevalence of pathogens) of production and harvesting zones;

    On-line information on monitoring of key biological and environmental parameters;

    Up to date information on methods used in disease detection in order to facilitate the understanding of laboratory results by producers and competent authority;

    Up to date information on current production practices and innovations.

- Existing communication platforms for shellfish safety could be employed to host the above-mentioned information on shellfish health.

BENEFITS :

- Increased stakeholders' awareness regarding bivalve diseases and requirements relating to shellfish health.

- Enhanced knowledge and commitment to health surveillance and management systems.

- Real-time alert communications on disease outbreaks.

- Improved production by better disease prevention and reduction of pathogen spread.

- Opportunities for using information systems to cover the whole value chain, in a one-health system.

THE MAIN LIMITATIONS :

- There are few established platforms for stakeholders and hence the cost of setting up operational communication networks will be expensive.

- Will require promotion of new platform and will require buy-in from producers.

*see the definition in the glossary

3. FACILITATING CRISIS MANAGEMENT

This is primarily the responsibility of the competent authority in very close collaboration with the shellfish industry, and diagnostic laboratories.

DESCRIPTION :

- The introduction of pathogens into a non-infected country or zone or the emergence of a new pathogen/genotype/strain may result in high mortality. **Before such events occur** and in order to decrease losses, the competent authority should develop a **contingency plan***. The competent authority should designate an operational unit involving representatives of key stakeholders that would be in charge of the co-ordination of all control measures to be carried out in emergency situation.

- The Competent Authority should identify all the staff required in case of crisis, including diagnostic laboratories and specify their responsabilities.

- Conditions which require contingency plans to be mobilised should be clearly specified and may include:

    Introduction of certain pathogens that would need to be listed at regional or EU level;
    Emergence of a new pathogen or a new genotype/strain;
    A mortality threshold combined with a geographic extent of the outbreak.

- The following protocols need to be established and shared :

    Protocols for handling/disposal of bivalves;
    Protocols to decrease the risk of pathogen spread and development at the local level;
    Protocols to establish quarantine and observation (surveillance*) zones;
    Protocols to control movements of bivalves;
    Reporting procedures;
    Diagnostic procedures.

- Communication and decision-making pathways must be clearly defined (see also recommendations 1 and 2) :

All the staff with specified roles in the contingency plan must understand their role and be prepared to act quickly to assist in the implementation of the plan.

This preparedness may be achieved through conducting periodic reviews of the plan with members of the operational unit and identification of training requirements / shortfalls to ensure that skills in field, administrative and diagnostic procedures are maintained for relevant staff.

BENEFITS :

- Shared and ready to be applied **protocols for crisis management**:

    Protocols with clear communication and decision-making pathways;

    Rapid and effective response capacity in critical periods;

    Rapid and effective progress in obtaining solutions and results bringing benefits to the production of molluscs;

    A good communication may result in a better acceptance of management measures.

THE MAIN LIMITATIONS :

- Good understanding of technical issues is required to achieve good communication and consensus on decisions. Thus, training is a critical part for this to work properly.

- Risk of lack of consensus about the measures to implement, due to conflicts of interests.

- If protocols implementation means increased cost, farmers may be reluctant to adopt it.

- There are already some established platforms involving different stakeholders (farmers, competent authorities) which have not been efficient or sufficiently active.

*see the definition in the glossary

4. LESSON-LEARNING FROM PAST DISEASE OUTBREAKS

This recommendation requires collaboration between the competent authorities, diagnostic laboratories, researchers, farmers and hatcheries* as well as other relevant support agencies involved in working with the shellfish industry.

DESCRIPTION :

- Gather and share information on disease events through collaborative meetings with all relevant stakeholders.

- This approach aims to:

  Identify potential sources of pathogen introduction and spread and to identify ways to prevent further introductions and spread of this and other emerging diseases;

  Identify commonalities and differences in experiences relating to the factors that may have contributed to or exacerbated mortality in order to develop mitigation strategies;

  Identify areas requiring further investigation or research and establish relevant research programmes;

- Conduct periodic reviews to gather further information on the success of mitigation strategies and to update stakeholders on progress of ongoing research activities.

- Publish relevant data in an easily accessible format to make them available to all stakeholders.

BENEFITS :

- Fluid exchange of information between the relevant stakeholders involved in control of diseases, researchers and industry.

- Identification of things to do / not to do in the event of recurrence of the same disease.

- Development of mitigation strategies against the introduction and spread of this disease as well as potentially new / emerging diseases.

- Documentation to support the decision process.

THE MAIN LIMITATIONS :

- The fragmentation of the current research framework, with short term funding.

- Lesson-learning from past disease outbreaks is not possible without synthesis and continuity.

- The lack of a communication structure to allow information flow and establish links within a European committee aimed at fighting against mollusc diseases.

- Costs, even if they would be relatively low.

*see the definition in the glossary

GOVERNANCE ISSUES

Sampling campaign in Alfacs Bay, Delta del Ebro.
Picture : ©VIVALDI

Identifying zone status

Oyster farms in Brittany, France.
Picture: ©IFREMER

5. IMPROVING SURVEILLANCE* AND DETERMINING ZONE STATUS USING RISK-BASED AND SPREAD MODELS

This is primarily the responsibility of the competent authority working in collaboration with the industry.

DESCRIPTION :

- Use **risk-based and hydrodynamic models** to combine all relevant information to establish the geographic limits of zones free of disease.

- Implement a **range of surveillance* activities** depending on the circumstances of the zone:

  If observable mortality is unlikely due to the pathogen of interest and the host species present (e.g. infection with *Bonamia exitiosa* in *Ostrea edulis*), then **targeted active surveillance*** is required to maintain a free status.

  Risk based surveillance (RBS)* methods should be used to identify high risk farms and locations within the zones (using criteria such as proximity to depuration plants and live animal movements).

  Other surveillance approaches should be considered, including **use of sentinel animals***, e.g. in areas where infection in farmed species is expected to exist only at low levels and without observable signs.

BENEFITS :

- Ensure a more efficient use of resources.

- Ensure that the geographic limits of zones are more likely to retain a disease free status.

- Ensure that any pathogen incursion is rapidly detected, facilitating action to be taken for pathogen **containment*** or pathogen **elimination***.

- Cover known pathogens but also new and **emerging pathogens***.

THE MAIN LIMITATIONS :

- Risk-based and hydrodynamic models and active surveillance* are costly and require technical expertise:

  To construct the models and keep them updated;
  To obtain the data parameters needed to feed the models.

- Efforts need to ensure that farmers are aware of the benefits to secure their cooperation.

*see the definition in the glossary

Acting on animal movements

Movement of mussels, Ireland.
Picture: ©I. Arzul

6- AVOID BIVALVE TRANSFERS* PRESENTING A RISK TO SPREAD PATHOGENS (NON-REGULATED PATHOGENS)

This is primarily the responsibility of the competent authority working in collaboration with the shellfish industry.

DESCRIPTION :

- Implement **a code of conduct for pathogens of local importance**. The steps to be taken would be as follows :

  Compile the **list of pathogens** of local importance;

  Define “free” and “infected” zones* for pathogens of interest;

  A system for **controlling & recording transfers*** must be in place, or put in place, by the competent authority and the shellfish industry;

  Commitment must be reached **at a local level**, between **all** producers in any given zone, that they will follow the code of conduct;

  Accord on any **testing schedule** going forward and on the terms which will be applied to control the disease, such as **transfer* restrictions**. Agreement must be reached between the National Reference Laboratory, competent authority, producers and producer organisations.

- Co-construction of the code of conduct, sign off by all parties.

- Annually, results of testing should be evaluated and meetings held to ensure continued by-in to the code of conduct and discuss any changes required based on testing findings, disease impact etc.

BENEFITS :

- Reducing the risk of spreading pathogens/diseases.
- Maintaining health status of areas.
- Protecting disease/pathogen free stocks.

THE MAIN LIMITATIONS :

- Involving 100% of the producers within a zone might be difficult.
- For some pathogens, efficient diagnostic tools and information needed to define the sampling strategy might not be available.
- A regular update of zone status is necessary.
- Free trade within the EU might raise difficulties.

*see the definition in the glossary

7- MINIMIZING THE SOURCE OF PATHOGENS BASED ON EARLY DETECTION.

This is primarily the responsibility of the competent authority working in collaboration with the industry.

DESCRIPTION :

- The detection of **emerging*** and **endemic*** pathogens should lead to action by both producers and the competent authority to limit their spread.
- Early detection of pathogen is crucial for an effective response.
- Early detection currently relies on alert from farmers and other stakeholders (e.g. research institutes that monitor pathogens) to the competent authority when there is suspicion of disease or abnormal mortalities.
- A communication procedure is crucial for efficient reporting and alert.
- Early detection is useful if immediate action is possible (e.g. the establishment of **infected zones*** and restriction of transfers* of live animals).
- Continuous automated sampling and analysis of **environmental DNA** (eDNA*) may contribute to early detection of pathogens in the future.

BENEFITS :

- Limit the spread of pathogens through immediate action.
- Ultimately, reduce production losses and increase profitability.
- Provide better knowledge of marine diseases.
- Reinforce the bases for decision-making (e.g. establishment of areas free of listed pathogens).

THE MAIN LIMITATIONS :

- Continuous monitoring of **environmental DNA** (eDNA*) may have some limitations. The detection of eDNA does not necessarily mean that viable pathogen is present in sufficient concentrations to cause infection. A positive eDNA can, therefore, only be treated as suspicion which requires confirmation by other means (e.g culture or PCR).
- Current methods of disease detection generally require complementary tests before action is taken. Complementary tests include histology to demonstrate that there is an infection and PCR/ sequencing to assess pathogen identity.
- Depending on the number of suspicions arising from farmer reporting or continuous environmental monitoring, the resources (financial, human, physical) required for investigations may be high.
- Other - mainly epidemiological - information is needed to define infected zones*. Notably the geographic distribution of the pathogen and susceptible populations and hydrodynamic information on currents is needed to delineate infected zones*. In many areas, such data may be sparse.

*see the definition in the glossary

Mortality reporting

Ropes with oysters in Alfacs Bay, Delta del Ebro, Spain.
Picture: ©VIVALDI

8- DEVELOP A HARMONIZED METHOD TO EVALUATE MOLLUSC MORTALITY AT THE EU LEVEL

This is primarily the responsibility of the competent authority working in collaboration with the shellfish industry.

DESCRIPTION :

- Although under EU aquatic animal health legislation there is an obligation for farmers, veterinarians and others to report unexplained or increased mortality events, there are currently no standard, harmonized protocols at EU level for estimating mortality levels. It is therefore difficult to compare mortality between regions, countries or production systems*.

- It is recommended that an **ad hoc stakeholder-working group**, including epidemiologists, diagnosticians, producers and administrators, to be appointed to:

  Develop and propose protocols for estimating mortality levels:

  - protocols are needed to assess current* mortality (daily) and cumulative* mortality;

  - protocols should be supported with standard reporting forms to record i) background information (site, species, production method, environmental parameters) and ii) mortality details (by date, life-stage*, location etc.);

  Review existing mortality data and propose, based in part on expert opinion, default mortality thresholds* depending on species, life-stages* and production systems* for the reporting of abnormal mortality.

- To improve chances of uptake and effective use of the protocols, **training** farmers, their technicians and advisors is needed and should focus on:

  The use of standardized mortality recording protocols;

  The importance of rapid reporting of abnormal mortality to the competent authority.

BENEFITS :

- Standardized mortality estimation will provide consistent data.

- The method to estimate mortality can be periodically updated based on data generated through standardized reporting.

- Mortality data will allow epidemiological studies to make comparisons between regions, Member States and production systems*, and thus improve the evidence base for research to support aquatic animal health management.

THE MAIN LIMITATIONS :

- Achieving widespread adoption by farmers of standardized mortality recording would require:

  Training of farmers, their technicians and advisors in the use of protocols;

  Provision of incentives to farmers for reporting.

*see the definition in the glossary

9- IMPROVING REACTIVITY, SENSITIVITY AND STANDARDIZATION OF REPORTING AND INVESTIGATING MORTALITY

This is primarily the responsibility of the competent authority working in collaboration with the shellfish industry.

DESCRIPTION :

- **Mortality report and investigation** include (see also recommendation 8) :
    - Monitoring shellfish mortalities;
    - Sampling animals according to standardised protocols;
    - Analyses of mortality data combined to other data sources (environment, climate, sample results...);
    - Deviation from expected mortality pattern triggers warning and investigation.
- It should:
    - Be standardized at the EU level for different shellfish species and production systems;
    - Use clear, simple, rapid and user-friendly procedures for easy web-based data entry to report mortality;
    - Provide information feedback to reporters.
- **Monitoring** should be implemented at geographic scales larger than the individual farm (epidemiological zone e.g. bay).

BENEFITS :

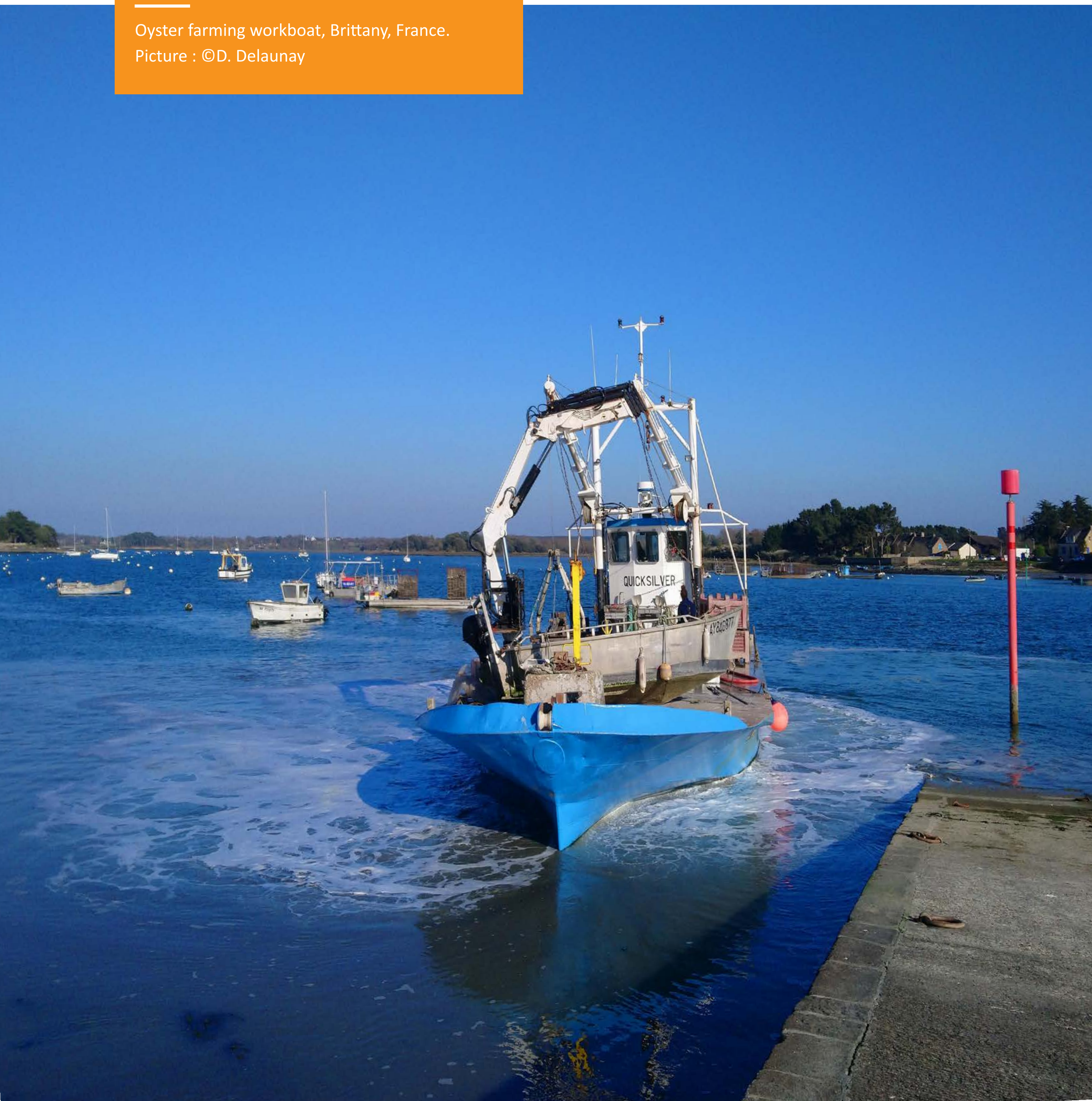
- Help **early detection** of emerging pathogens*.
- Help implementing faster response / mitigation measures to **prevent spread.**
- Provide open access data on the average and range of mortality at EU level and comparable data between countries.

THE MAIN LIMITATIONS :

- Data related:
    - Mortality estimation is not straightforward (difficulty to define the number of dead animals or an abnormal mortality - see also recommendation 8);
    - Some species or production systems* are difficult to monitor (e.g. deep beds).
- Data operating system related:
    - Development of web-based data entry may be expensive;
    - Open access to data is not guaranteed on a real-time basis;
    - Data sources and databases need to be sustainable.
- Stakeholder related:
    - Competent authority: must be able to investigate elevated mortalities and implement pre-defined action when appropriate;
    - Producers: must understand the reporting objectives and be committed to reporting.

*see the definition in the glossary

TECHNICAL ISSUES



Oyster farming workboat, Brittany, France.
Picture : ©D. Delaunay

Animal selection

Local oyster nursery, Ireland.
Picture : ©I. Arzul

10- DEVELOPMENT OF BREEDING PROGRAMMES* TO IMPROVE DISEASE RESISTANCE* FOLLOWING GOOD PRACTICES FOR PRODUCTION

This is the responsibility of hatcheries* working in collaboration with the industry.

DESCRIPTION :

- The goal of genetic selection is to **improve performances** of domesticated population according to the needs or demands. Genetic progress generated by selection must enable production and spread of spat with lower susceptibility to diseases.

- This improvement is achieved, generation after generation, by selecting and reproducing best individuals using internationally recognized and validated selection methods. Minimum requirements have to be followed to implement **sustainable** and **efficient breeding programmes***:

    Initial and characterized **genetic variability***;

    Breeding **goals and methods to measure traits** to be selected. Knowledge on **genetic parameters*** can help to quantify expected progress;

    Selection method (mass selection, family or sib selection, marker assisted selection, genomic selection) according to the traits to improve and the potential of financial investment;

    Selection pressure* to be applied by traits;

    Traceability process needs to be implemented;

    An acceptable increase of inbreeding rate* by generation to **manage inbreeding risks** and to preserve selection potential;

    Adapted facilities and in-house skilled human resources;

    Strategy to distribute genetic progress.

BENEFITS :

- Improve hatchery practices and management of inbreeding risk.

- Develop new products (spats) to improve survival in the field and growth benefits.

- Manage sanitary risk at the top of production chain in order to secure growing activity.

THE MAIN LIMITATIONS :

- Limited skilled human resources in hatchery practices and application of quantitative genetics (genetic selection).

- Limited studies and knowledge in genetic determinisms for resistance, production trait and abilities to perform in all environments (Genetic x Environment interactions).

- Need for protocols for field and control challenges to measure resistance or infection for commercial breeding programmes*.

- Lack of knowledge on feasibility for selection in polyploids.

- Limited genomic resources, efficient tools (robust QTL*, genetic markers for parentage assignment, high density and low density SNP* array for genomic selection), and feasibility studies to develop genetic selection and facilitate investment in genomic selection.

- Estimation of benefits and economic cost to implement efficient breeding programme*.

*see the definition in the glossary

11- FARMING OF SPAT SELECTED FOR LOWER SUSCEPTIBILITY TO DISEASES

This is the responsibility of farmers.

DESCRIPTION :

- Hatcheries* implement breeding programmes* to meet the needs of producers: limitation of susceptibility to diseases is one of the traits of interest.
- The evolution of technologies from mass selection to genomic selection using genetic markers should allow a balance to be reached between desirable traits such as resistance* to diseases and growth, yield, shell shape and color.
- Trade-off with other traits and maintaining the genetic diversity need to be carefully considered.
- Each producer should choose, purchase and rear spat on the basis of their own experience by benchmarking stocks against each other.

BENEFITS :

- Decreased mortalities as a result of lower susceptibility of oysters to diseases.
- Reduced pathogen spread resulting from lower infection intensity in populations.
- Decreased production costs due to lower stock losses through mortality.

THE MAIN LIMITATIONS :

- Potential selection of undesirable genetic traits such as susceptibility to other diseases as a result of genetic linkage with desirable traits genetic correlations.
- Genetic progress created by the hatchery may also be limited if rearing practices are not adapted or sub-optimal.
- The number of case studies using commercial populations to demonstrate genetic gain is still limited because genetic selection is still in its infancy in molluscs.

*see the definition in the glossary

Treating water

Depuration plant.
Picture: ©C. Aguilera

12- WATER TREATMENT MEASURES FOR LAND-BASED SHELLFISH SYSTEMS

This is primarily the responsibility of the industry (hatcheries*, expedition centers* and depuration plants*), who needs to work in collaboration with the competent authority (for regulatory issues), and with Research & Development institution/teams (for technological developments).

DESCRIPTION :

- **Shellfish land-based facilities** (Hatcheries*, Nurseries*, Expedition Centers* or Shellfish Depuration Plants*) can pose a **threat** when placed close to production sites if their effluents are not properly treated to prevent the spread of pathogens or bivalve life-stages*. Moreover, the inflow water into these facilities can also be a risk if it is not treated to prevent reinfection or pathogen spread.

- **Improved biosecurity** in shellfish land-based facilities, by means of **water treatments**, should be put in place routinely. Water treatment is a powerful tool for preventing not only recontamination of the stock, but also mortalities and the spread of pathogens into and out of a facility.

- Water treatments can be installed both in **flow-through*** or **closed systems***, such as recirculation aquaculture systems (**RAS**). The technical choice depends on the type of facility and the characteristics of the site (water source quality, location regarding shellfish production sites, etc.), ensuring that shellfish physiological activity is kept at the optimum.

- Several water treatments are regularly used for water disinfection/pathogen inactivation mainly: **ultraviolet light** (UV), **chlorine** and **ozone**. For biosecurity purposes, the treatment systems chosen should guarantee that the inflow water is safe for the stocks and the outflow water is free of both pathogens and phytoplankton, as well as shellfish life-stages*, which could propagate in open waters. In the case of nurseries*, the treatments should be less costly due to the large volumes of water needed (water sedimentation, solar UV, etc.).

- The technical characteristics of the water treatment systems can be very different; however, regardless of the choice of the system, their **efficient functioning needs to be assured**. Therefore, standard operating procedures need to be implemented, together with regular staff training, efficient monitoring programmes, and data recording/traceability, in order to guarantee the correct maintenance of installations.

BENEFITS :

- Ensure pathogen disinfection/inactivation (for shellfish and humans).

- Avoid pathogen entry into and out of land-based shellfish facilities.

- Allow specific pathogen-free status for shellfish hatcheries* placed close to positive production areas and/or in positive natural beds.

THE MAIN LIMITATIONS :

- Each facility should have its own biosecurity plan, which would have different stringent levels depending on the context.

- The effectiveness of the treatment is difficult to check, given that it is difficult to check the presence of pathogens in outflow water.

- High investment costs.

- Additional running costs and staff training.

*see the definition in the glossary

Elaborating technical recommendations based on geographic and species specificities

Lanterns used for oyster growing on long-line farm.
Picture: ©L. Gennari

13- ESTABLISH A CULTIVATION CALENDAR

This is primarily the responsibility of the industry (hatcheries*, nurseries* & farmers) and needs support from the competent authority and Research & Development institutions.

DESCRIPTION :

- An **immersion calendar** indicates the most suitable periods for the deployment of shellfish (spat introduction and stock movements) in terms of survival. These periods depend on host (species and physiological conditions), environment (temperature, hydrodynamics, food), production system* and pathogens (type, occurrence).
- An immersion calendar has to be site and species specific.
- An immersion calendar will rely on knowledge about pathogen occurrence and seasonality as to avoid deployment during periods of infection or mortality.
- For example, an immersion calendar for *Crassostrea gigas* exposed to OsHV-1 would be to deploy the oldest (biggest) animals as possible, to do it when temperature is well below 16°C (during the fall or the winter) and to adopt densities as low as possible according to production system*.

BENEFITS :

- Improve disease mitigation and survival.
- Assist zone management.
- Enforce cooperation between farmers.

THE MAIN LIMITATIONS :

- Establishing an immersion calendar relies on in-depth knowledge of the dynamics of pathogens in the environment and related data availability, but regular data gathering and monitoring may not be available everywhere.
- As an immersion calendar is site, pathogen and host specific, no generalization can be made.
- Immersion calendar needs to be compatible with spat supplies from hatchery or natural collection and with other zoo-technical or economic constraints.
- As producer's knowledge concerning production systems* and on-site information are fundamental and complementary to scientific and epidemiological information, collaborative approach based on transparency and openness will be a key factor for success.

*see the definition in the glossary

14- BIOSECURITY AND GOOD FARMING PRACTICES

The formal and legal basis of farming practices is the responsibility of the competent authority but expertise in relevant research and educational / training institutions as well as the industry are indispensable.

DESCRIPTION :

- Good farming practices can limit the spread of pathogens and mitigate their impact by improving survival, yield, and consequently, the economic viability of companies.
- Good farming practices comprise the use of **optimal techniques** for a particular location considering immersion time (bottom culture, intertidal culture, suspended culture) and culture support (cemented onto ropes vs. held in bags, lanterns, in baskets).
- For a specific cultivation system, good farming practices will also concern:

  Husbandry and handling practices;

  Spat origin and spat ploïdy as well as the calendar for spat introduction (see also recommendation 13).
- Mortality usually increases with the **stocking density** of the host and decreases with seawater renewal, due to a dilution effect of pathogens.
- The following recommendations can be made:

  Increasing density can increase disease transmission, but also reduce growth and host metabolism. Therefore, stocking density should be maintained in order to limit pathogen dispersion and disease risk.

  In intertidal areas, **rearing height** may be adapted to reduce infection through lowering exposure to pathogen, while maintaining acceptable growth rate. Farming conditions or handling should not be changed during an epidemic or a stressful period (see also recommendation 15).

  Animals should be **acclimated to new conditions** (cultivation structure, density or height) avoiding handling stress, abrupt changes in water conditions (temperature) and pathogen exposure.

BENEFITS :

- Higher survival of farmed stocks.
- Better growth and quality performance.
- Reduced spreading of pathogens.

THE MAIN LIMITATIONS :

- Industrial cost/ benefit might be reduced or compromised at short term.
- Strategies are site and species specific, which makes it challenging to apply to small-scale productions areas.
- Farmers might be reluctant to change/adapt their farming practices, to invest or reduce their production capacities if economic sustainability is not guaranteed.

15- TEMPERATURE MANAGEMENT IN CULTIVATION, HANDLING AND HARVESTING PRACTICES FOR *CRASSOSTREA GIGAS*

This is primarily the responsibility of the industry

DESCRIPTION :

- Adjust farming practices (immersion time, harvesting time, stock manipulation) avoiding or taking into account **critical periods**, when temperatures are favourable for the pathogen expression.

- Protect against spat losses in areas where OsHV-1µVar is endemic* by:

  Taking into account the temperature profiles in the production area to plan when *C. gigas* spat should be immersed avoiding temperatures conducive to viral replication (16-24ºC);

  Keeping spat cool during transport from the hatchery to the site;

  Ensuring that transfer* times are kept to a minimum;

  Avoiding oyster transfers* during high risk temperature periods.

- In areas where *Vibrio aestuarianus* is associated with recurrent mortality:

  Avoid handling (turning & grading) of oysters in periods of extreme heat;

  Consider hand grading where and when feasible;

  Use water graders instead of traditional graders or use water bins for the oysters to fall into at high impact points in traditional graders;

  Return stock to the water as soon as possible after grading;

  Following grading & hardening immerse oysters in cold water tanks prior to shipping;

  Differences between harvesting, transport and immersion temperatures should be managed to minimise stress on the oysters especially for movements of oysters during periods of warm weather when the disease is active.

BENEFITS :

- Potential reduction in number and amplitude of outbreaks of OsHV-1µVar in *Crassostrea gigas* spat.

- Reduction of mortality in older oysters associated with the presence of *Vibrio aestuarianus* in the summer months.

THE MAIN LIMITATIONS :

- The protocol improvement will depend on the country /region and will need to be established through trial and error.

- Efforts are required to ensure industry is aware that a good transport and immersion programme can be beneficial in terms of increased production.

- Rapid growth in the high risk period often means that oysters need to be handled to maintain shape or because the bags become too heavy which may also adversely affect oyster survival. Pre-planning by reducing densities prior to the summer may alleviate this issue.

- There could be potential difficulties/ incompatibilities between the “ideal” husbandry protocol to avoid critical temperature periods and the established culture practices at the site. The established practices may be tied to the market requirements of the business.

*see the definition in the glossary

Adapting farming practices and structures

Oyster spat collected in Brittany, France.
Picture: ©S. Pouvreau

16- DEVELOP LOCAL PRODUCTION SYSTEMS*

This is primarily the responsibility of the producers themselves or a group of producers (cooperative).

It requires the support of the competent authority working in collaboration with the industry.

DESCRIPTION :

- Develop **local sources** of spat supply from hatcheries*, nurseries* or collection sites
- **Reduce** frequency of **stock movements** during a production cycle.

BENEFITS :

- Reduce transfers* of live animals.
- Reduce risk of introduction of disease or non-native species.
- Reduce disease spread.
- Increase local adaptation and greater genetic diversity.
- Reduce transport costs.
- Reduce the carbon footprint of growers.

THE MAIN LIMITATIONS :

- Require administrative formalities
- Ensure the profitability of new operations, need for market research.
- Encourage producers to work collectively by sourcing locally.
- Benefits of breeding for disease resistance* cannot be easily achieved at small and local scales.
- Regulation may be required to encourage use of locally produced spat.

*see the definition in the glossary

17- DISPOSAL OF DEAD ANIMALS

This is primarily the responsibility of the producers themselves or a group of producers (cooperative). It requires the support of the competent authority working in collaboration with the industry.

DESCRIPTION :

- Evacuate the **dead animals** to eliminate a source of infection:

  extract them out of the natural environment or of the **closed system*** (hatchery, depuration centre, stocking tank in farms...)

  they **should always be treated and not translocated** anywhere.

- Treat the remains to avoid the dissemination of pathogens, in accordance with the national legislation (incineration, composting...). In case of a regulated disease occurrence, specific destinations may be mandatory defined for an appropriate management.

- This operation of sub-products' management should be described in the **biosecurity plan**.

BENEFITS :

- Avoid the spread of diseases.
- Assist in fighting and controlling diseases in an infected environment.

THE MAIN LIMITATIONS :

- Early detection tools are needed:

  to detect the disease before the infected organic matter is already wide spread in the environment;

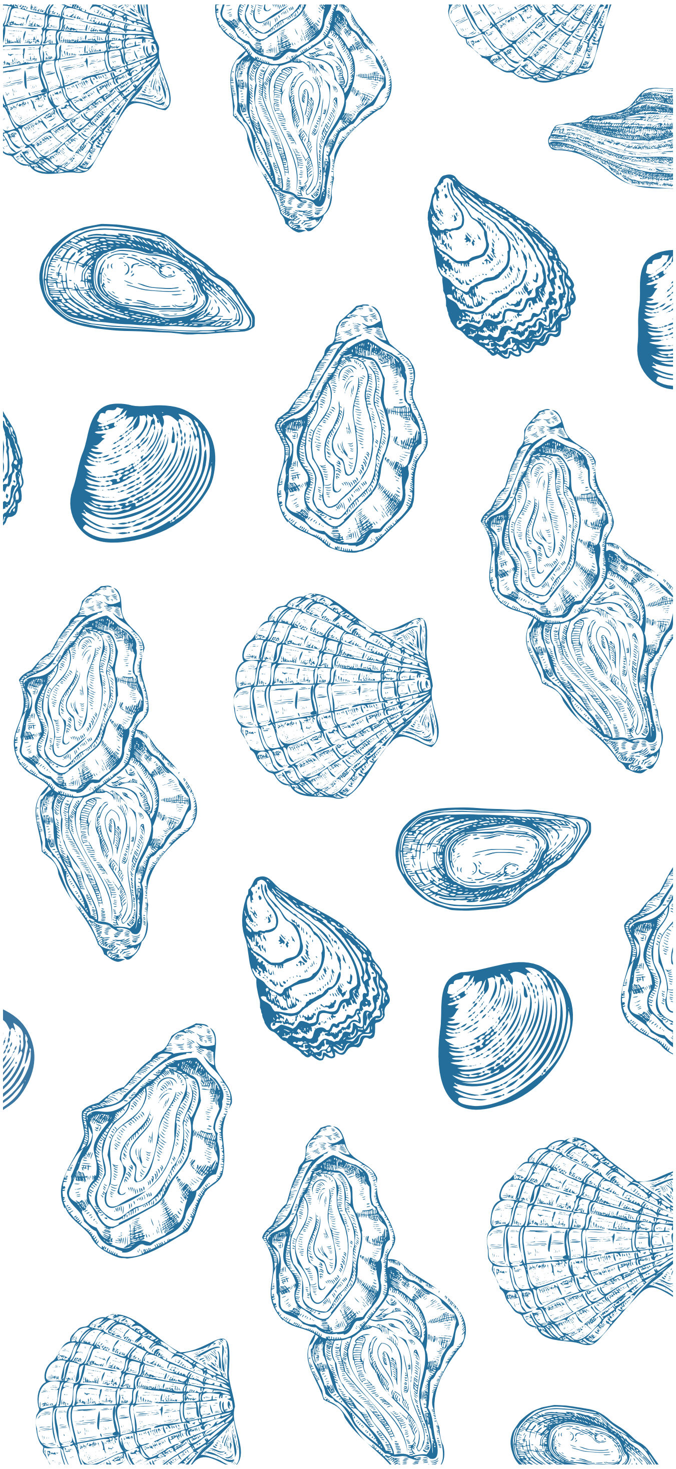
  to limit the extent of the elimination of the whole batch, including live animals, in late discovered cases.

- Detection tools are pathogen specific.
- Elimination and destruction methods to separate living animals from the dead are time consuming and costly.
- Training is required for farmers, e.g. using best practice and/or guidance on the management of sub-products.

*see the definition in the glossary

Oyster farms in Brittany, France.
Picture: ©IFREMER

CONCLUSION



Many scientific studies have been carried out in VIVALDI and the results obtained in the different work packages (https://www.vivaldi-project.eu/) have allowed identifying a set of recommendations.
Already existing guidelines, from the different countries involved in this work, were studied and considered for the compilation of this set of recommendations.

Some recommendations that would need further research to become effective have not been included in the Manual, but might be of interest in the future, including the stimulation of bivalve immunity, the diversification of farmed species or the introduction of natural barriers between culture areas.

The co-construction methodology used to identify, describe, organize and review the recommendations included in VIVALDI deliverable D6.10 and to elaborate the manual presented some challenges and benefits.
Gathering different stakeholders around the same table requires flexibility and organization, especially when they come from different countries and speak different languages. Alternating sub-group discussions (in native language) and plenary gatherings (in English with the support of the scientists) was an answer to this difficulty.
This methodology has encouraged fruitful exchanges between scientists, producers and competent authorities. It contributed to developing a better understanding, acceptance and sense of ownership of disease management measures by the concerned stakeholders.
This approach has set the path to a structured cooperation among the shellfish industry stakeholders and could be shared and used in other contexts for example to support the evolution and implementation of the legislation across Europe.

The result of this work is a set of 17 recommendations included in this manual for disease management and biosecurity, a friendly and easy-to use tool for producers and competent authorities.

Mussel rafts in Galicia, Spain.
Picture: ©I. Arzul

GLOSSARY

Breeding programmes
Breeding programmes are the planned breeding of a group of animals or plants, usually involving at least several individuals and extending over several generations. Breeding programmes are set up with the aim to exploit genetic variation in a sustainable way.

Closed system
Aquaculture facility where water is recirculated and, usually, treated (oxygenated, disinfected and temperature regulated), to improve its quality for stock holding and safety for the environment before its discharge.

Contingency plan
Work plan describing actions, requirements and resources (including human resources) needed to control and eventually eradicate a disease.

Current (daily) mortality and cumulative mortality
Daily mortality is the number of animals dying in a 24 hour period. Cumulative mortality is the number of dead individuals over a fixed period. For example, if 10,000 oysters are stocked at one point of time and 5,000 are harvested 12 months later, by difference the cumulative mortality for that period is 50%.

Disease resistance /tolerance
Resistance is the ability of the host to limit pathogen burden whereas tolerance is the ability to limit the disease severity induced by a given pathogen burden.

Emerging pathogen / Endemic pathogen
An emerging pathogen is a previously unknown microorganism infecting bivalves or a previously known pathogen infecting a new bivalve host species, exhibiting a different pathology (e.g. increased virulence), or rapidly expanding its geographic range into new locations, e.g. transboundary spread. In contrast, an endemic pathogen is a pathogen constantly present in a population.

Environmental DNA (eDNA)
Environmental DNA or eDNA means DNA extracted from environmental samples including water or sediment without prior isolation of any targeted organism. This DNA includes DNA from cells or live organisms, extracellular DNA coming from degraded or dead cells.

Expedition centre/dispatch centre
Logistic wet facility for preparation of shellfish for the distribution chain.

Flow-through system
Continuous water flow aquaculture facilit y with neither reused nor retention of the water that passes through, and is directly discharge after its use.

Genetic parameters
Heritability and genetic correlation are genetic parameters which describe possibilities for selection. Heritability of a trait is the part of phenotypic variability explained by the genetic resemblance between individuals from the population, it measures the ability of parents to pass on their capacity for a trait to their offspring. Genetic correlation quantifies the genetic relation between two traits.

Genetic variability
Genetic variability describes the variety of genes in the population. This parameter has to be considered in the long-term management of population to avoid potential deleterious impacts of inbreeding, conserve adaptative capacities and maintain accuracy of breeding values in genetic evaluations.

Hatcheries
Establishments hosting the shellfish reproduction phase in controlled conditions. Hatcheries generally include dedicated rooms for the storage and maturation of broodstock and for the breeding of larvae.

Inbreeding rate
Inbreeding rate is the results from the mating between related individuals. Inbreeding rate provides the probability that offspring receives identical genes from both parents. Inbreeding could generate deleterious effects on traits.

Infected zone/Free zone
An infected zone is a defined geographic area in one or more countries where the mollusc population is known to be infected with a specific pathogen based on the results of a surveillance programme.
In contrast, in a disease-free zone, surveillance, biosecurity and control measures have been applied to demonstrate and maintain freedom from one or more specified pathogens.

Life-stages
A life stage for farmed molluscs may be determined by the lifecycle of the species and the production system. Life stages include fertilised eggs, free-swimming larvae, shelled larvae, followed by various stages of adult development which vary between species and production systems but are generally measured by shell size.

Mortality thresholds
A mortality threshold is the level of mortality above which action must be taken. The mortality threshold is most likely to be monitored as a cumulative mortality over a period of one or two weeks. The action may be, for example, reporting to the competent authority, or a decision to harvest.

Nurseries
Establishments hosting early on-growing phase in shellfish production, starting from the fixation of larvae.

Pathogen containment
Containment means that pathogen spread is limited geographically to a defined geographic area through the application of control and biosecurity measures (creating a zone with a specific health status). Containment will prevent an increase in prevalence and mitigate the impact of the disease on susceptible populations.

Pathogen elimination
Pathogen elimination is the reduction in prevalence of an infectious pathogen or parasite in a defined geographic area (e.g. country, region) to zero.

Production systems
A molluscan production system is defined by: i) species or mixture of species, ii) environment (marine, brackish, freshwater), iii) product (for human consumption or on-growing), iv) water source (open water or controlled, i.e. in a hatchery), and by iv) infrastructure e.g. ponds, lines or ropes, rafts, bags or none.

Quantitative Trait Loci (QTL)
Section of DNA in the genome which is closely correlated with a trait in a population.

Risk based surveillance (RBS)
Risk based surveillance is a type of targeted active surveillance where sections of the population are selected based on the level of risk of becoming infected with a specific pathogen, and developing clinical signs. This approach to surveillance requires estimates of the level of risk of a farm or farming areas in the surveillance programme of being infected and spreading pathogens. These estimates may be based on knowledge of animal movements, density of farms, connections via water, ...

Selection pressure
Selection pressure is the ratio of the number of selected individuals to the total number of candidate individuals. The smaller this ratio, the greater the pressure and the better the efficiency is.

Sentinel animals
Sentinel animals might be externally sourced and are known to be susceptible to the pathogen targeted by the surveillance programme and highly likely to develop clinical signs if infected. Monitoring sentinel animals should provide advance warning of the presence of the pathogen in a farm or farming area.

Shellfish depuration plant
Wet facility for removing microbiological contamination from harvested shellfish, prior human consumption, operating with clean (natural or disinfected) sea water.

SNP
Variation in the genome of a species (polymorphism) at a single base pair.

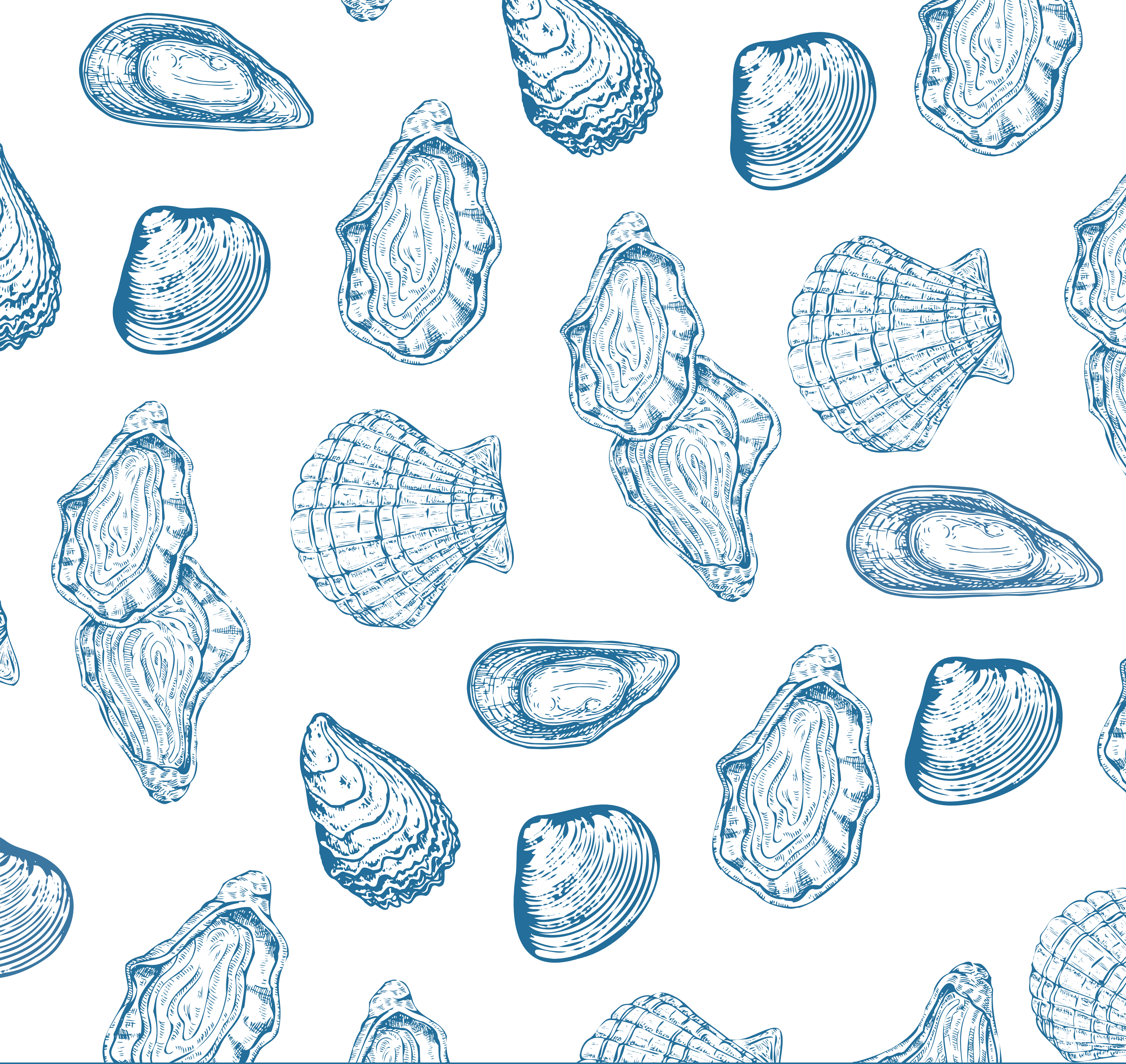
Surveillance
The systematic, continuous or repeated, measurement, collection, collation, analysis, interpretation and timely dissemination of animal health and welfare related data from defined populations.

Targeted active surveillance
Active surveillance implies programed sampling, following structured protocols. In contrast, passive surveillance depends on reports/ alerts, such mortalities, from stakeholders or citizens.
Targeted active surveillance means looking for a specific pathogen (or other pre-defined hazards) in selected sections of the bivalve population. Such surveillance optimizes the effort decreasing resources required (in contrast to general surveillance which is not specific to one or more pathogens; syndromic surveillance – collecting information on signs of disease - is a type of general surveillance).

Transfer (of animals)
Transfer of animals is the intentional movement of animals by transport.

Probe to monitor environmental parameters in oyster and mussel farming area.
Picture: ©VIVALDI

Blue mussels *Mytilus edulis*.
Picture: ©I. Arzul



This project has received funding from the European Union's Horizon 2020 Research and innovation programme under grant agreement N° 678589