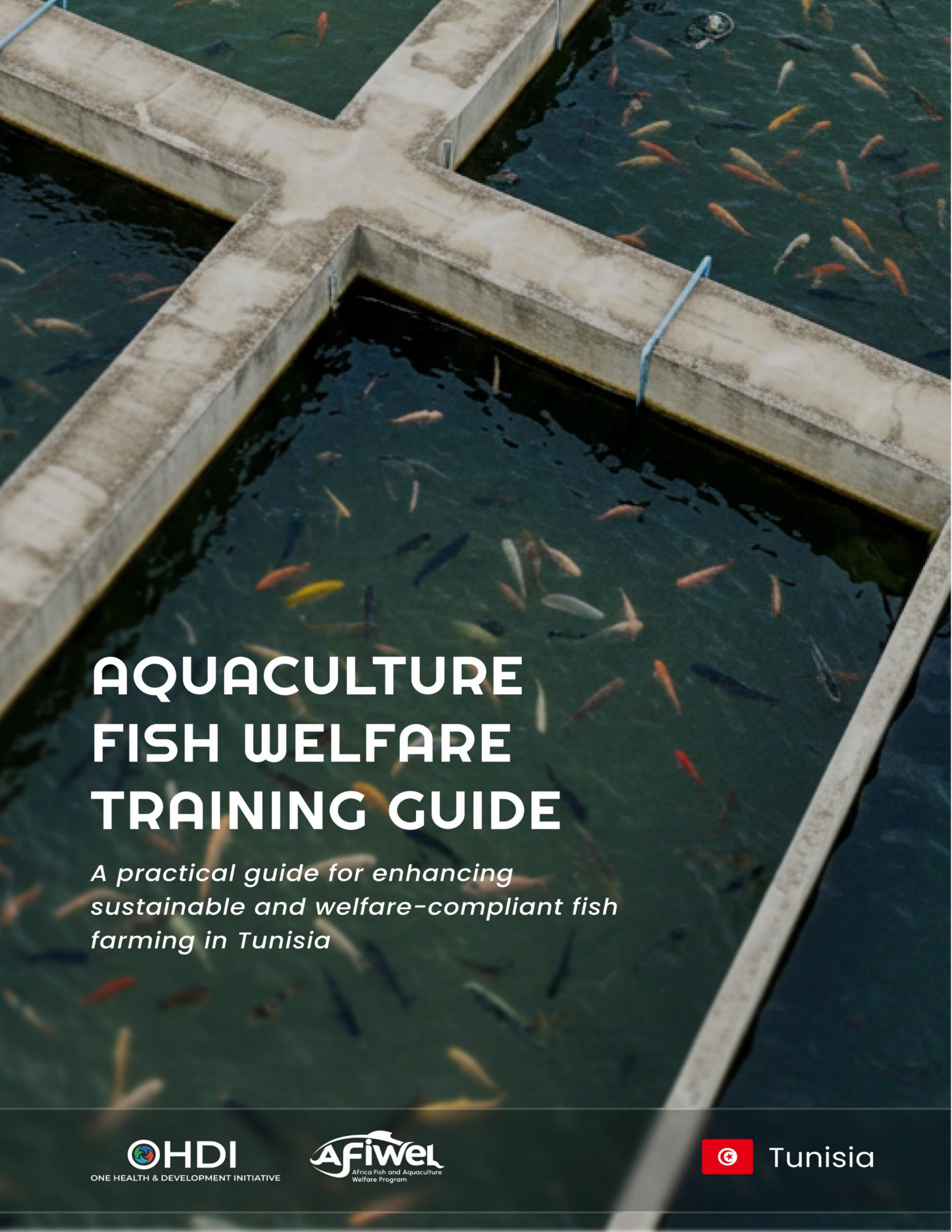

AQUACULTURE FISH WELFARE TRAINING GUIDE

A practical guide for enhancing sustainable and welfare-compliant fish farming in Tunisia

OHDI
ONE HEALTH & DEVELOPMENT INITIATIVE

AFIWEL
Africa Fish and Aquaculture Welfare Program

Tunisia

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ABREVIATIONS AND ACRONYMS

CTA: Centre Technique d'Aquaculture

DGPA: Direction Générale de Pêche et d'Aquaculture

FAO: Food and Agriculture Organisation of the United Nations

FAWC: Farm Animal Welfare Council

GIPP: Groupement Interprofessionnel des Produits de la Pêche

INSTM: Institut National des Sciences et Technologies de la Mer

ONP: Office National des Pêches

RSPCA: Royal Society for the Prevention of Cruelty to Animals

SDG: Sustainable Development Goals

WOAH: World Organisation for Animal Health

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PREFACE

Fish welfare is increasingly recognised as a key element of sustainable and ethical aquaculture. In Africa, where aquaculture is essential for food security, livelihoods, and economic progress, there is a rising urgency to integrate welfare principles into production systems, policy frameworks, and capacity-building initiatives.

The Africa Fish and Aquaculture Welfare (AFIWEL) Program, implemented by One Health and Development Initiative (OHDI), was established to address this need. The AFIWEL program is a pan-African initiative that supports ethical, welfare-driven, safe and sustainable aquatic life and production systems across Africa. One of its flagship initiatives is the AFIWEL Fellowship, which engages select fisheries and aquaculture professionals and experts in capacity-building, community-building and field implementation programs to advance fish and aquaculture welfare practices and integrate them into existing sustainable aquaculture frameworks. Through this pan-African fellowship model, the program supports professionals across the continent in leading transformative action for fish and aquaculture welfare through education, stakeholder engagement, and policy advocacy.

This Fish Welfare Training Guide is one of several developed by AFIWEL Fellows. This particular guide has been tailored to the specific aquaculture realities of Tunisia, providing practical, evidence-based knowledge and tools for fish farmers, aquaculture workers, extension officers, animal health professionals, and institutions involved in the fish production value chain.

The content draws from global best practices, scientific insights, and local expertise to ensure that welfare recommendations are both technically sound and contextually relevant. It covers key aspects such as water quality, stocking densities, feeding, handling, transportation, health management, and humane slaughter, all anchored in the principles of good welfare practices: freedom from pain, distress, discomfort, and suffering.

As you explore this guide, we invite you to reflect on the broader goal it serves, which is to promote responsible aquaculture systems that protect animal welfare, support livelihoods, and ensure long-term environmental sustainability. We hope it will be a valuable resource in your efforts to improve fish health, welfare, productivity and sustainability outcomes in Tunisia and across Africa.

With best regards,

The AFIWEL Program Team

One Health and Development Initiative (OHDI)

MODULE 1 – OVERVIEW OF AQUACULTURE IN TUNISIA

What is Aquaculture?

Aquaculture is the farming of aquatic organisms, such as fish, crustaceans, molluscs and algae, in controlled environments. It aims to produce aquatic products for human consumption, industry, or the repopulation of wild populations. Unlike fishing, which involves capturing aquatic organisms in their natural habitat, aquaculture involves raising them in tanks, ponds, floating cages at sea or other artificial systems.

Fishing and Aquaculture in Tunisia

With its two seafronts running along 1,350 km, a national maritime domain of 80,000 km², and 105,200 hectares of lagoons, Tunisia has always been a country of sailors, and fishing has always been an activity of considerable importance. This strategic sector accounts for 8% of the value of agricultural production and 1.1% of the gross national product, generating approximately 53,000 direct jobs. The port infrastructure comprises 40 ports, including ten offshore ports: Tabarka, Bizerte, La Goulette, Kelibia, Sousse, Teboulba, Mahdia, Sfax, Gabes, and Zarzis, as well as 30 coastal ports and landing sites. The total capacity of these ports is 150,000 tonnes of seafood per year (www.ctaqua.tn). The fishing fleet comprises approximately 12,265 units, with 91% of these being coastal boats. The fishing and aquaculture sector offers 43,011 jobs, distributed as follows: 73% in coastal fishing, 11% in light fishing, 11% in trawling, 3% in shore fishing, and 2% in aquaculture (DGPA, 2023).

The volume of seafood exports in Tunisia fluctuates around 37,062 tonnes for a value close to 846 million dinars, thus placing it in second place in exports of agricultural and agri-food products after olive oil (DGPA, 2023). Around 75% of exports are directed to EU markets. The main exported products are cephalopods (octopus and cuttlefish), crustaceans (shrimp and prawns), shellfish, and fresh fish (including bluefin tuna), as well as aquaculture products. The development strategy for the fisheries sector is based on the preservation of benthic resources, the exploitation of small pelagic resources, the enhancement of the added value of commercial fishing products, and the development of aquaculture.

Brief Overview of Aquaculture in Tunisia

Historical

Aquaculture in Tunisia is an ancient activity dating back to Roman times, as evidenced by the mosaics in the Bardo Museum in Tunis. The recent Tunisian experience in this field dates back to the 1960s. Initiated by the private sector, it began with the breeding of the Mediterranean mussel *Mytilus galloprovincialis* and the Pacific oyster *Crassostrea gigas* on fixed tables in Bizerte (Figure 1).

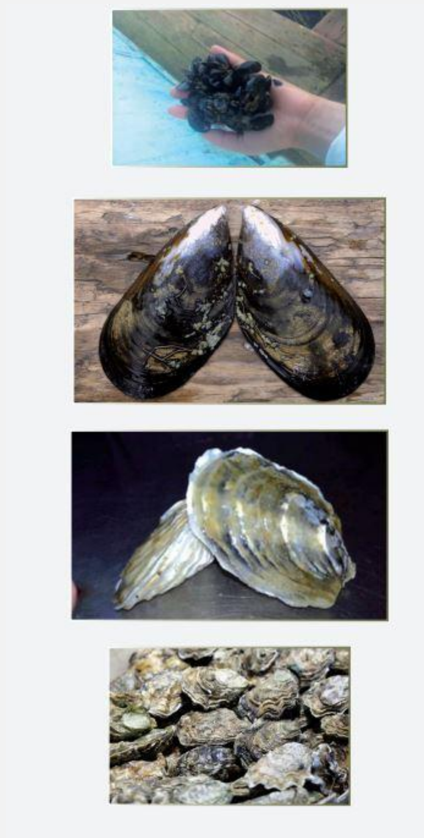

Figure 1 Pacific oyster, Crassostrea gigas, on fixed tables in Bizerte

The supply of mussel spat is done locally by capture in the Bizerte lagoon, while that of oyster is done by importing from abroad (France, Italy, etc.). Subsequently, the shellfish farming facilities were transferred to the National Fisheries Office (ONP), which continued these activities and began the construction of ponds in the lagoons of Monastir and Tunis and began jointly with INSTOP (National Scientific and Technical Institute of Oceanography and Fisheries, currently INSTM) the stocking of certain dam reservoirs with fry of various species and their exploitation by fishing (common carp; bighead mullet; pig mullet, etc.).

In the early 1980s, one of the first private hatcheries for sea bass (*Dicentrarchus labrax*) and sea bream (*Sparus aurata*) in the Mediterranean was established in the south of the country by private operators supported by banks in the region (Figure 2).

Figure 2 Sea bream (Sparus aurata) and Sea bass (Dicentrarchus labrax)(www.gipp.tn)

The 1990s were marked by the implementation of the aquaculture master plan and the development of continental fish farming in inland freshwater bodies in extensive mode, although with some timid private achievements, which mainly concerned the land-based farming of sea bass and sea bream and shellfish farming in the Bizerte lagoon (fixed tables and floating lines) following the liquidation of the ONP.

Since 2003, a new aquaculture activity has emerged, making an exceptional leap in terms of the adoption of new farming techniques: the fattening of bluefin tuna *Thunnus thynnus* which not only ensures a weight gain of more than 20% in a few months, but also allows the sale of this product on the international market at times when the tuna flesh is fatty with the best prices. The tuna, from fishing and intended for fattening, is transferred alive to floating cages in the open sea. It is fattened there in captivity for a few months before being sold fresh at relatively more profitable prices.

Recent years have seen the expansion of floating and submersible cage farming of sea bass and sea bream. The supply of fry and feed is mainly through imports from abroad (France, Italy, etc.).

National strategy

From a strategic perspective, the Tunisian government has implemented two ten-year aquaculture development strategies: the Aquaculture Master Plan (1996–2006) and the National Strategy for the Development of Aquaculture (2007–2016). The objectives of this strategy are to ensure the sustainable development of aquaculture by:

- Improving the management of the aquaculture sector
- Quantitative and qualitative development of livestock products
- Increasing annual consumption per capita.

In 2019, the General Directorate of Fisheries and Aquaculture, in collaboration with sector stakeholders, developed a Strategic Study of Fisheries and Aquaculture for 2030 (www.ctaqua.tn). Based on a participatory approach, the study aims to advance the fisheries and aquaculture sector while ensuring the preservation and sustainable, rational use of fishery resources. It proposes a dynamic national action plan built around six strategic pillars, including the sustainable development of the aquaculture sector, whose main strategic objectives are:

- Improving the governance of the sector
- Improving the performance of aquaculture projects
- Strengthening scientific research in the sector

- Strengthening training in the sector
- Mastering health and zoo sanitary aspects
- Promoting the local market and encouraging exports
- Protecting the environment

Aquaculture Fish Production Systems in Tunisia

Aquaculture in Tunisia encompasses various breeding systems tailored to the country's geographical specificities and the species cultivated. Here are the main Tunisian aquaculture systems:

1. Mariculture (Sea farming)

The first marine fish farming initiative began in 1984 in the Boughrara lagoon south of the island of Djerba to produce 400 tonnes/year of sea bass (*Dicentrarchus labrax*) and sea bream (*Sparus aurata*) in floating cages. Several natural disasters caused by toxic phytoplankton blooms, such as Dinoflagellates in the Boughrara Sea, have destabilised the operation of the farm (Figure 3).

The second initiative was installed 50 km north of Sousse and designed to produce sea bass and sea bream in raceways located on land (www.ctaqua.tn).

Figure 3 Sea bass and sea bream farming (Gharbi, 2024)

Marine aquaculture provides approximately 1,200 direct jobs and a total of 2,000 jobs, and is experiencing a growing evolution in its share of the total production in the fisheries and aquaculture sector. This share has evolved significantly over the last ten financial years, reaching approximately 13% of total production in terms of quantity and 22% in value as of 2016 (www.gipp.tn).

2. Shellfish farming

Shellfish farms are dedicated to the farming of molluscs, particularly mussels and oysters. The Bizerte lagoon is a major site for these activities. Although the Tunisian experience in farming Mediterranean mussels and Pacific oysters dates back more than half a century, this sector has not been able to exceed a production of 200 tonnes/year. Estimates are always beyond actual production (www.ctaqua.tn).

3. Freshwater aquaculture

The Tunisian experience in exploiting dam reservoirs for fishing dates back to the 1960s. This activity was initiated by the National Fisheries Office (ONP) through the stocking of certain dam reservoirs with fry of various species and their exploitation by fishing. The promulgation of the decree by the Minister of Agriculture on 20

September 1994, relating to fishing in dams, courses and expanses of freshwater, made it possible to regulate and supervise this activity.

Currently, nine governorates are involved in this type of activity (Beja, Ben Arous, Bizerte, Jendouba, Le Kef, Nabeul, Zaghouan, Kairouan and Siliana). The number of boats is estimated to be 232, and the number of fishermen is 450. These fishermen are farmers from the interior regions of the country who have a fairly low cash level. Extensive fish farming offers them the opportunity to profitably produce cheap fish, which they can easily sell or consume (www.ctaqua.tn).

In inland areas, such as Siliana and Kairouan, artificial ponds and basins are used for farming freshwater fish, including carp and tilapia.

4. Aquaponics

Aquaponics combines fish farming and plant cultivation in a symbiotic relationship. In Tunisia, initiatives are emerging to promote this sustainable system. For example, the Facebook page "TunisieAquaponie" presents local projects in this field (www.tunisie-aquaponie.com) (Figure 4).

Figure 4 Aquaponic unit (www.ctaqua.tn)

5. Intensive and semi-intensive systems

Intensive systems involve high stocking density with strict control of environmental conditions, while semi-intensive systems combine natural and artificial methods to optimise production (Figures 5, 6, 7).

Figure 5 Floating cage (actual work, 2024)

Figure 6 Intensive breeding of Nile tilapia in the South of Tunisia (www.ctaqua.tn)

Figure 7 Intensive cultivation of spirulina(www.ctaqua.tn)

These systems reflect the diversity and dynamism of the Tunisian aquaculture sector, contributing to the country’s food security and economic development.

Q&A Session

- What are the major challenges encountered in the management of aquaculture systems in Tunisia, and what solutions are envisaged?
- How does Tunisia position itself in relation to other Mediterranean countries in terms of aquaculture production?
- What are the specific environmental impacts of floating cages at sea on local ecosystems?
- How do local communities perceive the impact of aquaculture on their daily lives?
- Is aquaculture a real need in the existing Tunisian fisheries system?

MODULE 2 – INTRODUCTION TO ANIMAL WELFARE

Overview, History and Trends of Animal Welfare

Animal welfare refers to how an animal perceives and responds to its environment. It encompasses both physical and mental aspects, assessing whether the animal's needs are being met (Broom, 2011). The five fundamental freedoms, defined by the Farm Animal Welfare Council (FAWC), provide a framework for evaluating animal welfare:

- Freedom from hunger and thirst: access to adequate food.
- Freedom from discomfort: suitable environmental conditions.
- Freedom from pain, injury and disease: appropriate veterinary care.
- Freedom to express natural behaviours: space and conditions that allow for normal behaviours.
- Freedom from fear and distress: protection from stress (Brambell, 1965)

History

Agricultural societies used animals primarily as work tools and food resources. However, some philosophical traditions, such as Buddhism and Jainism, emphasised compassion toward living beings (Fraser, 2008). In the Western world, ideas about the treatment of animals have been influenced by figures such as Aristotle, who saw animals as subordinate to humans (Fraser, 2008). René Descartes viewed animals as biological machines devoid of consciousness. This mechanistic view influenced the way animals were treated. In contrast, philosophers such as Jean-Jacques Rousseau and Jeremy Bentham argued for animal rights. Bentham asked the fundamental question: "Can they suffer?", emphasising that the capacity to feel pain should be the central criterion (Fraser, 2008).

The first animal welfare laws emerged in 1822, with the introduction of the Cruel Treatment of Cattle Act in the United Kingdom (Broom, 2011). In 1824, animal protection societies were established, such as the Royal Society for the Prevention of Cruelty to Animals (RSPCA) (Fraser, 2008). Following Brambell's report, the development of animal welfare science occurred in the 1960s, with studies of the behavioural and physiological needs of animals. In 1965, the Brambell Report introduced the basis of the "Five Freedoms" (Brambell, 1965). The World Organisation for Animal Health (WOAH) has integrated animal welfare into its standards since the 2000s (WOAH, 2021). Mellor and Beausoleil (2015) discussed the development of labels and certifications based on welfare standards (e.g. Organic Agriculture, Global Animal Partnership).

Trends

Science continues to improve our understanding of how animals experience pain, emotion, and stress. Advanced techniques, including brain imaging and physiological indicators, now allow for more objective assessments of well-being, while the integration of the "quality of life" concept complements traditional approaches (Mellor and Beausoleil, 2015). In livestock farming and agriculture, there has been a shift towards more welfare-friendly production systems, such as free-range and outdoor rearing (Broom, 2011).

For aquaculture, there was a development of sustainable aquaculture farming practices (e.g. fish welfare) and a reduction in antibiotics through better stress management. (WOAH, 2021). Fraser (2008) demonstrated the introduction of policies aimed at reducing suffering in wildlife management practices, highlighting a conflict between individual well-being and large-scale conservation. Mellor and Beausoleil (2015), in their study, they reveal the use of artificial intelligence and the Internet of Things (IoT) to monitor welfare parameters on farms. Additionally, Fraser (2008) demonstrated that the demand for ethical and animal welfare-friendly products is on the rise. The World Organisation for Animal Health (WOAH) is calling for the strengthening of national and international laws to improve the welfare of animals in farms, laboratories, and zoos, as well as the adoption of global strategies to integrate animal welfare into the Sustainable Development Goals (SDGs) (WOAH, 2021).

The Five Freedoms of Animal Welfare

The Five Freedoms of Animal Welfare are fundamental principles developed by the Farm Animal Welfare Council (FAWC) in 1965 and updated in 1979. They aim to ensure that the basic needs of animals are met, particularly in farms, zoos and other human-controlled environments.

1. **Freedom from hunger and thirst:** Animals must always have access to clean water and adequate feed that meets their physiological needs. This ensures healthy growth, efficient production (in the case of livestock), and the prevention of diseases related to malnutrition.
   Example: In cattle farms, an automatic waterer must be available to prevent dehydration, especially during periods of extreme heat.
   Impact: A lack of food or water can weaken an animal's immune system, increasing its vulnerability to disease.

2. **Freedom from discomfort:** Animals must be kept in an environment that provides them with appropriate shelter, clean resting areas, and optimal temperatures. This aspect also includes managing hygiene in their habitats to reduce the risk of disease.
   Example: Pigs should have a clean, non-slippery, dry floor with an area protected from the elements.
   Impact: An unsuitable environment can lead to injuries, heat stress, or infections.
3. **Freedom from pain, injury and disease**: Animals must receive prompt and appropriate veterinary care, as well as disease prevention through vaccination, disinfection and regular health monitoring.
   Example: In poultry farms, regular vaccination campaigns help prevent avian influenza, also known as bird flu.

Impact: This limits the physical suffering of animals and improves their quality of life and productivity.

4. **Freedom to express normal behaviours:** Animals must be allowed to behave naturally according to their species. This requires suitable facilities and social interaction with their peers where relevant.
   Example: Laying hens in a free-range system can scratch the ground and perch, natural behaviours not possible in cages.
   Impact: Behavioural restrictions increase stress, abnormal behaviours (such as pecking), and decrease overall well-being.
5. **Freedom from fear and stress:** Animals should not be subjected to violent treatment, stressful living conditions, or practices that generate excessive fear (for example, rough handling during transport).
   Example: Restraint techniques used to handle cattle should be gentle and reduce panic.
   Impact: Stressed animals produce hormones such as cortisol, which affect not only their health but also, in the case of farm animals, the quality of their products (meat, milk, etc.).
   The application of these principles is essential to promote animal welfare in animal farms, research laboratories, zoos, and even natural habitats. They also provide an ethical framework for judging human practices involving animals.

These freedoms can be achieved through specific management practices that are directly linked to each, such as access to nutritious feed and veterinary support, humane handling and slaughter, appropriate surroundings (and shelter) and management (Figure 8).

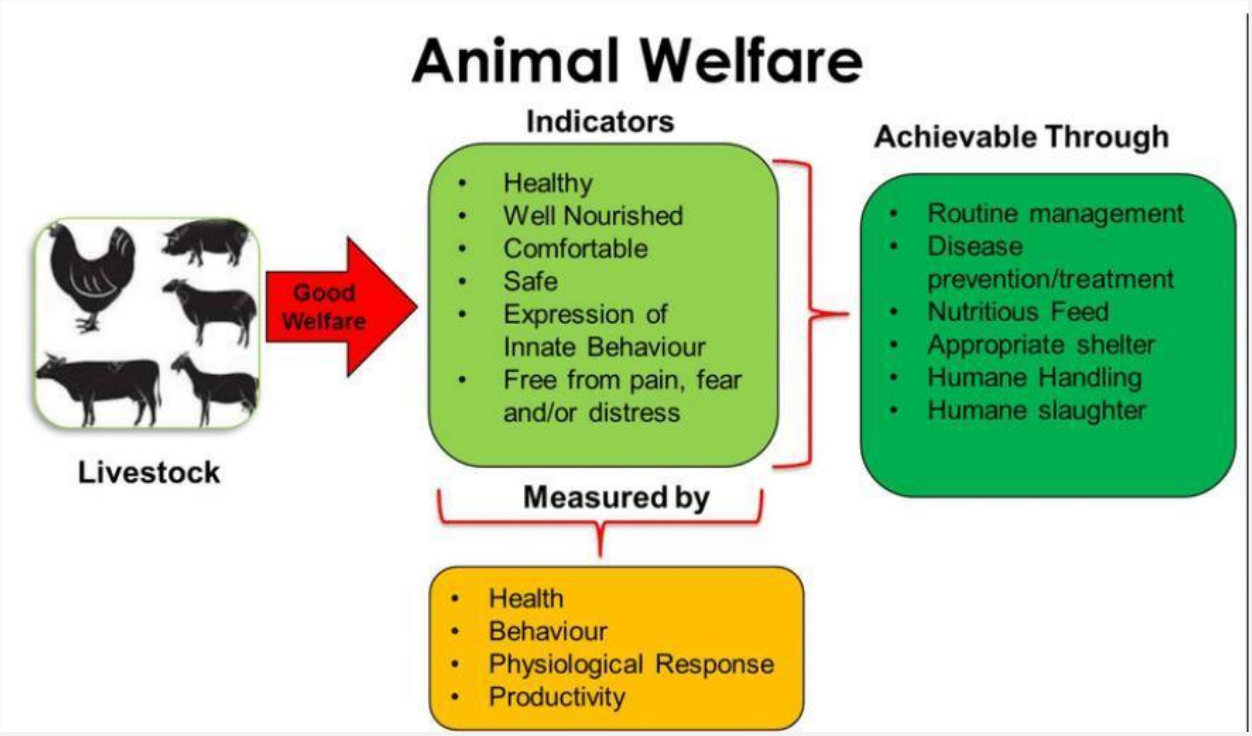

Figure 8 A summary of good welfare indicators, how to achieve them and how they can be quantified (Njisane et al, 2020)

Figure 9 illustrates the impact of various types of stress on production performance and efficiency, with the final consequences evident in the quality of the end product.

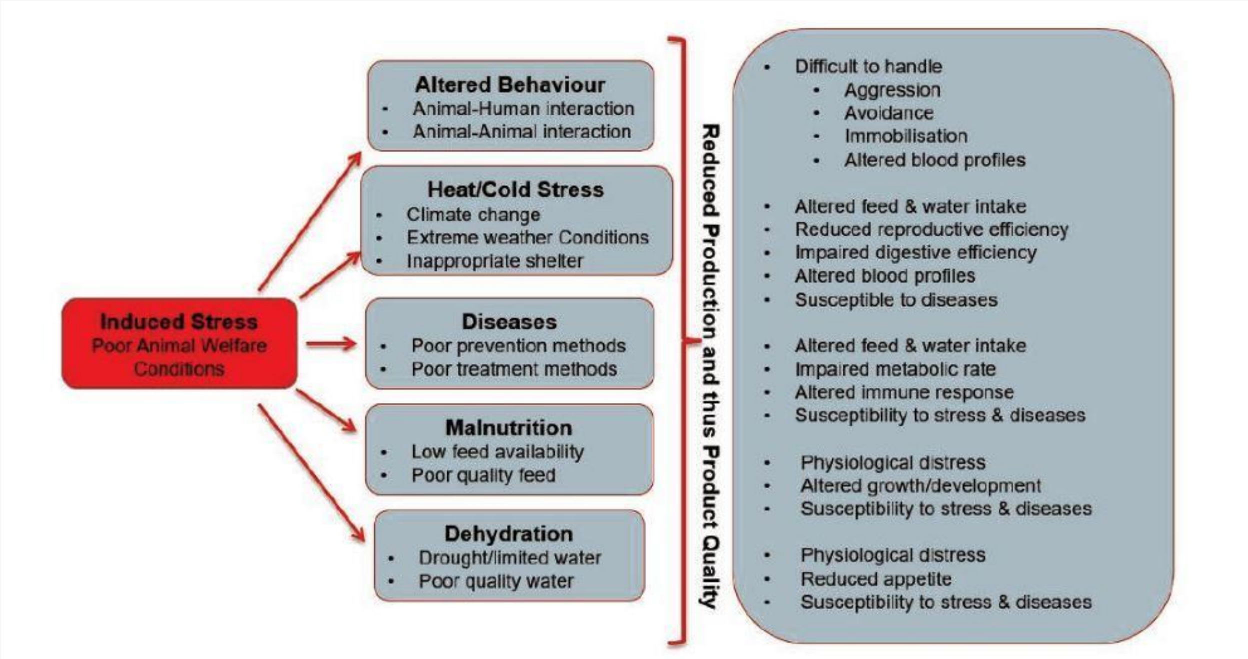

Figure 9 An illustration of poor animal welfare and stress indicators impacting on production and product quality (Njisane et al, 2020)

The Five Domains of Animal Welfare

The five domains of animal welfare are a more recent and comprehensive framework for assessing and improving animal welfare. Developed by Professor David Mellor and colleagues in the 1990s, these domains expand on the principles of the five freedoms by also considering animals' mental states, in addition to their physical needs.

1. **Nutrition (Physical Domain):** This domain concerns access to adequate nutrition and hydration to meet the physiological needs of animals.
   Examples: Ensuring animals receive a balanced diet, preventing both undernutrition and overnutrition, and providing clean, continuously available water.
   Impact: Reduced hunger, thirst and nutritional imbalances, promoting optimal health.
2. **Environment (Physical domain):** This domain includes physical living conditions, such as temperature, available space, and habitat safety.

Examples: Protect animals from extreme weather conditions by providing sufficient space for their movement and a clean resting place.

Impact: Avoid physical discomfort, injury, or heat stress, thereby supporting their overall well-being.

3. **Health (Physical domain):** This involves the prevention, diagnosis and treatment of disease, as well as the management of pain or injury.

   Examples: Regular vaccinations, appropriate veterinary treatments, and management of chronic diseases.

   Impact: Reduction of suffering, maintenance of good physical health and increased longevity.

4. **Behaviour (Functional Domain):** This field is concerned with the ability of animals to express natural behaviours and benefit from appropriate social interactions.

   Examples: Allowing animals to explore, play, forage for food and interact with their peers in an enriched environment.

   Impact: Reduction of frustration and abnormal behaviours, promoting a more fulfilling life.

5. **Mental state (Affective domain):** This field focuses on the emotional and cognitive experiences of animals, including positive sensations and the reduction of states of stress or fear.

   Examples: Avoid rough handling, reduce anxiety-provoking situations, and provide pleasant experiences such as play or petting (depending on the species).

   Impact: Promote positive emotional states such as comfort, security, and pleasure.

The first four areas (nutrition, environment, health and behaviour) directly influence the fifth, which is the animal's mental state. The goal is to ensure that animals not only live without suffering but also benefit from positive experiences in their daily lives.

The Five Freedoms and the Five Domains of Animal Welfare are two complementary frameworks that aim to ensure animal welfare, but they differ in their scope and level of detail (Table 1).

Table 1 The core differences between the Five Domains and the Five Freedoms Source: (Daisy Sopel, 2021)

The Five Freedoms	The Five Domains
1. Freedom from hunger and thirst	1. Nutrition - giving sufficient, balanced, varied, and clean food and water.
2. Freedom from discomfort	2. Environment - comfort through temperature, substrate, space, air, odour, noise, and predictability.
3. Freedom from pain, injury and disease	3. Health - enabling good health through the absence of disease, injury, impairment with a good fitness level.
4. Freedom to express normal behaviour	4. Behaviour - providing varied, novel, and engaging enrichment through sensory inputs, exploration, foraging, bonding, playing, retreating, and others.
5. Freedom from fear and distress	5. Mental state - the animal should benefit from predominantly positive states, e.g., pleasure or comfort, while reducing negative states such as fear, frustration, hunger, pain, or boredom.

Animal Welfare in African Conditions

Developed countries have a high sense of concern for farm animal welfare, which has been a rapidly growing area of interest over the years (Njisane and Muchenje, 2017). Regardless of some research-based recommendations that have been developed towards mitigating these concerns, some people generally perceive animal-based food consumption as an inhumane act (Muchenje *et al.*, 2018). While international animal welfare standards exist in the developed world, leading to improved management procedures, there are inherent factors that impede their adoption in most developing regions, such as Africa. There is a need to acknowledge the geographical, climatic and systematic differences between the developed and developing worlds (Njisane and Muchenje, 2017). Some communities are uncertain about and unfamiliar with the concept of animal welfare. Furthermore, there is limited research and published literature in this area, based on African communities and practices (Fraser, 2008). According to Ndou *et al.*, low priority is given to AW in the developing world, and this can be related to traditional customs and beliefs, a lack of knowledge in animal handling and sub-standard handling facilities.

Large numbers of smallholder farmers and pastoralists keep numerous livestock, which contribute significantly to the food production in Africa (Hoffman, 2010). Furthermore, small-scale farming plays an important role in the rural economy (Nyika, 2009). These are usually based in remote and/or rural areas, sometimes characterised by limited resources and access to some knowledge (Herrero, 2013).

Chulayo and Muchenje reported that animal welfare is generally associated with producers, retailers and the industry, with no consumer consideration, though it may affect their attitudes towards and purchase decisions of certain products. Hence, the current status regarding awareness of AW matters disqualifies the region from import and export participation with the rest of the world, as reflected in a sluggish contribution towards economic growth (Scholtz *et al.*, 2011).

Pastoral farming is another example of traditional practices found among some rural societies in East Africa (Gray *et al.*, 2003). Furthermore, Degen reported that about 70%, 50%, and 40% of the total land in Kenya, Tanzania and Uganda, respectively, is occupied by pastoralists. In this system, the herdsman moves from one place to another with the livestock on foot, in search of feed and water.

Some African cultures perform slaughter on animals in their conscious state (Clottey, 1985), paying little or no attention to following the suggested humane handling or slaughter practices (Grandin, 2018).

Legal Framework for Animal and Fish Welfare in Tunisia

In Tunisia, the legal framework regarding animal welfare is still in its infancy compared to that of some European countries. However, some legislative texts govern the protection of animals, particularly in agricultural and health contexts:

- The Tunisian Penal Code criminalises acts of cruelty to animals, but the penalties remain relatively light.
- Law No. 2005-95 indirectly mentions the management of animals on farms and slaughterhouses.
- Law No. 2019-25 of February 26, 2019, relating to the health safety of foodstuffs and animal feed.

These regulations remain limited in terms of detail and strict enforcement. Ongoing reforms aim to strengthen animal welfare standards, particularly under the influence of international organisations.

According to WOAH, the Tunisian agricultural sector, particularly livestock farming, plays a vital role in the economy. Animal welfare practices are often influenced by:

- The pastoral tradition, which gives a certain consideration to animals, particularly in terms of access to water and grazing conditions.
- Intensive livestock farming infrastructure, often associated with animal welfare issues such as overcrowding, inadequate slaughter conditions and heat stress, especially in summer.

MODULE 3 – INTRODUCTION TO FISH WELFARE

What is Fish Welfare?

It represents the "State of physical and biological balance where fish do not exhibit disease or excessive stress" (Conte, 2004). Fish should be able to swim, hide, forage for food and interact socially as they would in the wild (Brown *et al.*, 2011).

Main Factors Influencing Fish Welfare

- **Water quality:** Parameters such as dissolved oxygen, temperature, pH and ammonia must be maintained at optimum levels (FAO).
- **Stocking density:** Overcrowding can cause stress, increase disease risk and restrict movement (WOAH).
- **Feeding:** Fish require adequate feed quantity and quality to meet their energy and nutritional needs (Ashley, 2007).
- **Handling and transport:** Practices should minimise stress and injury (RSPCA).
- **Disease prevention:** Rigorous health monitoring reduces the risk of spreading diseases (Huntingford *et al.*, 2006).

Benefits of Improved Aquaculture Fish Welfare

The implementation of better protection of fish in aquaculture offers many benefits, both for the fish themselves, for producers, and for consumers. These benefits cover ethical, economic, environmental and product quality aspects.

1. **Improved fish welfare:** Healthy fish, living in an environment that respects their biological and behavioural needs, have less stress, less disease, and reduced mortality. This promotes the expression of natural behaviours, contributing to more respectful animal husbandry (Ashley, 2007).
2. **Increased productivity:** Good fish health allows for faster growth and better feed conversion, thereby reducing production costs. Reducing losses due to disease or poor conditions improves yields (Huntingford *et al.*, 2006).

3. **High quality of aquaculture products:** Fish raised under optimal conditions produce better quality meat, with improved texture, flavour and nutritional composition. Consumers are increasingly favouring products from ethical and sustainable aquaculture (Conte, 2004).
4. **Reduced costs related to illness:** Better protection limits the occurrence of pathologies, which reduces the costs associated with treatments (antibiotics, vaccines) and economic losses due to mortality. Example: Biosecurity and health protocols help prevent epidemics (FAO, 2020).
5. **Compliance with regulations and certifications:** Meeting animal welfare standards helps meet legal requirements and benefit from certifications (such as ASC, Global GAP). These labels increase the value of products in international markets. Example: The Aquaculture Stewardship Council (ASC) certification includes welfare criteria for fish (WOAH, 2021).
6. **Environmental sustainability:** Systems that protect fish often involve efficient management of resources (water, feed, energy), thereby reducing environmental impact. Healthy fish require fewer antibiotics, which reduces the risk of antimicrobial resistance and pollution (FAO, 2020).
7. **Improving the perception of aquaculture:** Ethical and welfare-focused aquaculture attracts public support and builds consumer trust in the industry. Example: Animal welfare awareness campaigns contribute to transparency and social acceptance of aquaculture practices (RSPCA, 2021).

Introduction to Fish Welfare Practices

Fish welfare has become a major concern in the aquaculture, fisheries and scientific research sectors. Fish, as sensitive vertebrates, experience pain, stress and other forms of distress, which calls for responsible and ethical management of their living conditions. In aquaculture, fish welfare is directly linked to their health, growth and the quality of the products produced by this industry (Huntingford *et al.*, 2006).

Fish welfare practices involve providing an environment that meets the physiological, behavioural, and social needs of fish. This includes water quality control, disease prevention, adequate stocking density, and appropriate feeding. These measures are not only beneficial to the fish but also essential for ensuring sustainable and profitable production (Ashley, 2007).

Adopting welfare-focused practices not only helps meet consumer expectations for ethics and sustainability but also helps comply with increasingly stringent international animal welfare regulations (WOAH, 2021).

Fish Welfare in Tunisia

Tunisia, as a member of the OIE, is gradually adopting international standards in animal welfare. Tunisia is also concerned with the welfare of marine animals, particularly in connection with fishing and aquaculture (FAO):

- **Artisanal fishing:** It can have indirect impacts on protected marine species, such as sea turtles. Small-scale fishing represents a large part of fishing activity in Tunisia, but practices can cause significant stress to fish:
    - Long delays between capture and killing, leading to prolonged suffering.
    - Use of nets that cause injuries and accidental mortality for non-target species (turtles, dolphins).

In industrial fishing, the conditions of large-scale capture, such as trawling, exacerbate stress and physical damage.

- **Aquaculture:** Awareness of ethical practices in fish farming is beginning to emerge, although the concept of marine animal welfare remains relatively new.

Aquaculture in Tunisia is booming, particularly for species such as sea bass, sea bream and Bluefin tuna. However, several aspects pose challenges to the welfare of the fish:

a) Breeding conditions

- Excessive density: Fish are often raised in overcrowded cages or ponds, limiting their mobility and increasing their stress.
- Water quality: Pollution or poor management of environmental parameters can lead to disease and mass mortality.

b) Handling and transport

- Transport: Fish undergo considerable stress during transfer between farms and markets, often without adequate protective measures.
- Vaccination and treatments: Handling during treatments can be rough and prolong their suffering.

c) Slaughter

- Slaughter methods remain rudimentary, with little use of techniques such as electro-anaesthesia or rapid cooling, which could reduce suffering.

FAO is collaborating with Tunisia to enhance the sustainability of aquaculture systems and eliminate harmful practices. In Tunisia, laws specific to fish welfare are still limited and in their early stages of development. However, several legislative texts and indirect regulations govern aspects related to fish protection, mainly in the areas of fisheries, aquaculture, and marine conservation. Here is an overview of the relevant laws and regulations:

- Law No. 94-13 of 31 January 1994 relating to the exercise of maritime fishing: This law governs the exercise of maritime fishing, including the management of fishery resources and the preservation of marine ecosystems. It prohibits certain destructive practices (such as fishing with explosives or toxic substances) that can inflict unnecessary suffering on fish. It establishes periods of biological rest to allow reproduction and preserve populations.

  Limitations: It does not explicitly address aspects related to the suffering or welfare of captured fish.

- Decree No. 94-1744 of 29 August 1994 establishing the conditions for exercising maritime fishing: This decree supplements Law No. 94-13 and establishes technical rules for maritime fishing, such as the types of fishing gear authorised. Banning certain fishing nets and techniques reduces destructive impacts on fish and non-target species.
  Limitations: Lack of specific clauses on practices that respect the welfare of fish during and after their capture.

- Law No. 2009-49 of 20 July 2009 relating to protected marine areas: This law aims to create and manage marine protected areas for the conservation of marine biodiversity. Protected areas provide refuges where fish can live and reproduce without the pressure of fishing. It indirectly contributes to fish welfare by limiting human disturbance in these areas.
- Law No. 2005-95 of 18 October 2005 relating to food safety: This law deals with health standards in food chains, including the production and processing of aquaculture fish. Indirectly, it encourages the maintenance of optimal breeding conditions to ensure the quality of the products, such as maintaining good water quality and providing adequate feed for the fish. However, it does not address the conditions of handling and slaughter.
- Law No. 2010-49 of 7 July 2010 on organic products: This law promotes organic production systems, including aquaculture. Organic farming imposes stricter standards for fish density in ponds and the use of environmentally friendly methods, which indirectly promotes the well-being of fish.

The welfare of aquaculture fish is gaining recognition in Tunisia, but specific laws are still being developed. Recent discussions on the adoption of international standards (notably from the WOAH) show a willingness to evolve.

Q&A Session

- How to raise awareness among fishermen and aquaculturists about the importance of fish welfare?
- What are the economic and ecological benefits of improving fish welfare in Tunisia?
- Why is the welfare of fish less taken into account than that of terrestrial animals in Tunisian policies?
- What international partnerships could help Tunisia strengthen its fish welfare practices and laws?
- What is the perception of local communities regarding fish welfare?
- Is the Tunisian consumer ready to pay a premium price for aquatic products that meet animal welfare standards?

MODULE 4 – GROWING SYSTEMS AND FISH WELFARE

Modern aquaculture relies on growth systems that optimise production while ensuring fish welfare. These systems, whether intensive or extensive, directly influence the health, growth, and quality of life of fish. Effective management is crucial to address the biological and ethical challenges inherent in fish farming.

Site Selection

Selecting an aquaculture site is a crucial step in planning an aquaculture project. It influences not only productivity, but also the sustainability of the activity and the welfare of the fish. A rigorous analysis of environmental, economic and social criteria is essential to ensure the success of the project while respecting the regulations in force.

1. **Environmental Criteria**

   a) Water quality

   Water quality is a crucial factor in maintaining fish health and promoting growth. Key parameters include:

   - Temperature: Should be appropriate to the needs of the species being farmed. For example, tilapia thrive at 25–30°C.
   - Dissolved oxygen: Adequate oxygen levels are crucial to avoid fish stress and mortality.
   - pH: Most aquaculture species prefer a pH between 6.5 and 8.5.
   - Salinity: Should be compatible with the species (fresh, brackish or marine).
   - Nutrient loading: Ammonia and nitrite levels should be controlled to avoid toxicity.

   Timmons and Ebeling (2010) emphasise the importance of constant monitoring of water quality to ensure animal welfare and productivity.

b) Topography and geology

The site should have a topography that allows for natural drainage and minimises excavation costs. The soil should be impermeable to prevent excessive water infiltration in ponds. Clay soils are ideal for pond systems. Boyd (1998) notes that soil composition affects water retention and maintenance costs.

c) Climate

Local climatic conditions (temperature, precipitation, wind, seasonality) must be adapted to the species being farmed. For example, tropical areas are ideal for species such as tilapia and shrimp. FAO (2020) indicates that climatic conditions determine production cycles and influence the profitability of aquaculture systems.

d) Availability and quality of water resources

A reliable source of water is essential, whether it comes from a river, lake, borehole, or the sea. Water must be available in sufficient quantities to meet livestock needs throughout the year. According to Bunting and Pretty (2007), the sustainability of aquaculture systems depends on access to a stable and quality water resource.

2. Socio-economic Criteria

a) Proximity to markets and infrastructure

The site should be located near roads, markets, or ports to facilitate the distribution of products. Infrastructure, such as electricity and processing facilities, should be accessible. Edwards (2000) explains that strategic location can reduce transportation costs and increase overall profitability.

b) Cost of land and legal access

The cost and availability of land should be compatible with the project budget. Verify that the site complies with local regulations, particularly those related to land use and environmental protection. FAO (2020) emphasises that legal compliance is a key element in avoiding litigation and ensuring the project's sustainability.

c) Social acceptance

Aquaculture can have social and environmental impacts. It is important to consult with local communities to ensure they support the project. Hambrey and Evans (2016) recommend a participatory approach to minimise conflict and maximise benefits for local people.

3. Risk Assessment

a) Environmental risks

Assess the risks of pollution of surrounding waters, soil erosion, or the introduction of invasive species. Boyd *et al.* (2020) remind us that aquaculture practices must respect the principles of environmental sustainability to avoid long-term negative impacts.

b) Climate risks

Identify risks associated with extreme weather events, including floods, droughts, and storms. Timmons and Ebeling (2010) advise choosing sites outside flood zones and planning mitigation measures.

c) Long-term economic viability

Analyse profitability projections taking into account initial costs and operating expenses. Bunting and Pretty (2007) emphasise the importance of feasibility studies to ensure a favourable return on investment.

Selecting an aquaculture site requires a multidimensional approach that takes into account environmental, social and economic criteria. A well-chosen site contributes not only to the economic success of the project but also to sustainability and compliance with animal welfare standards.

Rearing Systems

Aquaculture farming systems are diverse and tailored to meet specific objectives, depending on the species being farmed, available resources, and environmental conditions. Each system has advantages and challenges related to sustainability, fish welfare and productivity.

Here is a clear and structured summary table on the different aquaculture farming systems:

Table 2 Summary of different aquaculture farming systems

System type	Features	Benefits	Limits	References
Extensive system	- Little human intervention - Based on natural resources (food, oxygen, etc.)	- Low cost - Respects natural cycles	- Limited production - Vulnerability to environmental variations	Boyd et Tucker (1998)
Semi-intensive system	- Partial food supplements - Moderate control of environmental parameters	- Higher yield than extensive - Compatible with traditional practices	- Dependence on external resources - Pollution risks	FAO (2020)
Intensive system	- Strong human intervention (artificial feeding, advanced technology) - Strict control	- High efficiency - Precise control of environmental parameters	- High costs - Potential environmental impacts	Timmons et Ebeling (2010)
Pond system	- Use of natural or dug water bodies - Semi-natural environment	- Low cost - Suitable for hardy species (e.g. tilapia)	- Limited control of parameters (temperature, oxygen)	Boyd (1998)
Floating cage system	- Cages placed in natural bodies of water (fresh or marine water)	- Low installation cost - Efficient use of water bodies	- Exposure to predators	Conte (2004)

	- Exploitation of the existing environment		- Dependence on natural conditions	
RAS (Aquaculture Recirculation) System	- Closed system with filtration and water recycling - Continuous monitoring	- Reduced water usage - Full control of parameters	- High initial cost - Dependence on technological equipment	FAO (2020)
Aquaponics	- Integration of aquaculture and hydroponics - Nutrient recycling	- Joint production (fish + plants) - Resource efficiency	- Requires high technical expertise - Requires precise nutrient management	Rakocy *et al.* (2006)

Common Growing Facilities and Welfare Considerations

Culture facilities play a central role in aquaculture, directly influencing fish welfare. The design, management, and environmental parameters of these facilities are crucial for reducing stress, promoting fish health, and optimising production.

1. **Common Growing Facilities**

   - **The ponds:** Ponds are natural or artificial bodies of water suitable for hardy species such as tilapia and carp. They have the advantage of being inexpensive and compatible with natural cycles, but offer limited control over environmental parameters such as temperature or oxygen. In addition, they are vulnerable to climatic variations (Boyd, 1998) (Figure 10).

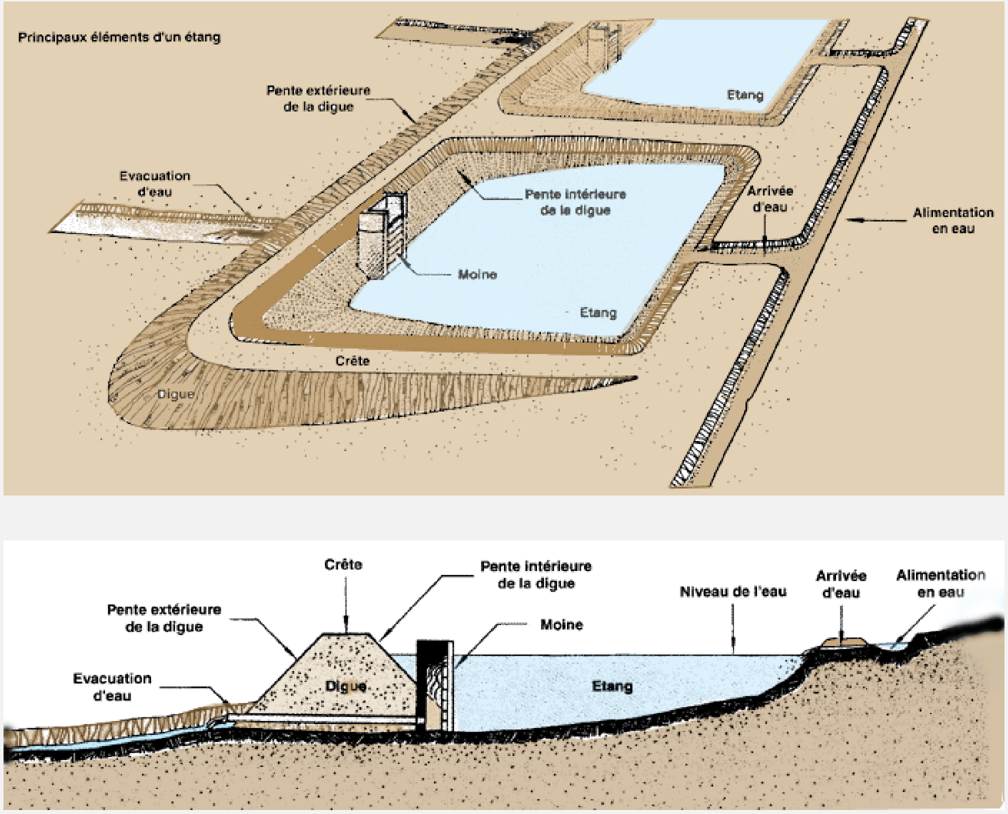

Figure 10 A fish pond (FAO)

- **Floating cages:** Floating cages are mesh structures installed in natural bodies of water such as lakes, rivers or the sea. They are often used for salmon and efficiently exploit natural aquatic resources. However, they are associated with increased risks of predation and sensitivity to environmental variations (Conte, 2004) (Figure 11).

Figure 11 Floating cage in Tunisia (ctaqua.tn)

- **Concrete basins:** Concrete ponds are artificial structures that allow precise control of water and fish. Their design facilitates easier harvesting and enables precise regulation of environmental conditions, although construction costs are high and they rely heavily on water management systems (FAO, 2020) (Figure 12).

Figure 12 Concrete basin at the Tilapia breeding station in Bechima, Tunisia (ctaqua.tn)

- **Recirculating systems (RAS):** Provide continuous water filtration and full control of environmental parameters. They reduce water usage and are compatible with high fish densities. However, these systems require high installation and maintenance costs, as well as technical expertise to manage failure risks (Timmons and Ebeling, 2010) (Figure 13).

Figure 13 RAS system in aquaculture (asc-aqua.org)

- **Raceway systems:** consist of elongated channels with a continuous flow of water, often used for cold-water species such as trout. They provide excellent water oxygenation and allow intensive production, but require a constant water flow and complex effluent management (Boyd and Tucker, 1998) (Figure 14).

Figure 14 Fish Raceway (aquaportail.com)

- **Aquaponics:** Aquaponics combines aquaculture and hydroponics, allowing dual production of fish and plants. This system has a low environmental impact, but it requires technical expertise and complex nutrient management (Rakocy *et al.*, 2006) (Figure 15).

Figure 15 Aquaponics System (ctaqua.tn)

2. Fish Welfare Considerations

Water quality is crucial, as inappropriate parameters, such as low oxygen levels, incorrect pH, or high ammonia levels, can cause stress and disease. Regular monitoring of these parameters and the use of effective filtration systems are recommended (Boyd and Tucker, 1998).

Stocking density also plays an important role. High densities can lead to stress, aggressive behaviour and rapid spread of disease. Therefore, it is essential to adapt the density to the species and the conditions of the facility (Huntingford *et al.*, 2006).

Water oxygenation is another key factor. Low oxygen levels can cause rapid mortality. The installation of suitable aerators or oxygenation systems is recommended (Timmons and Ebeling, 2010).

Feeding management is essential to avoid underfeeding or overfeeding, which can cause stress and metabolic imbalances. It is advisable to provide a balanced diet adapted to the needs of the species (FAO, 2020).

Handling of fish should be minimised to reduce injuries and stress. The use of suitable equipment is crucial to ensure their welfare (Conte, 2004).

Disease control relies on strict biosecurity practices, as well as responsible use of antibiotics and vaccines to limit the risks of spread (Huntingford *et al.*, 2006).

Finally, environmental enrichment, for example by adding structures that simulate the natural habitat, such as rocks or artificial vegetation, can improve welfare by reducing abnormal behaviours (Braithwaite *et al.*, 2006).

Stocking Density

Stocking density (kg fish/m^3) describes the biomass of fish per unit of water in the rearing system. Optimal stocking densities are based on several factors, such as the type of fish species, life stages, growing systems, and water flow rates, and can also depend on environmental conditions. It is also one of the main characteristics that determines whether a fish farm is extensive or intensive. It can have a major impact on fish welfare, as it influences water quality, growth, stress status and social interactions – such as aggression among the fish. For example, maintaining high water quality allows you to increase biomass or stocking density. However, if you don't maintain high water quality, you will need to lower stocking density; otherwise, this will lead to stress, and in extreme cases, death of fish.

For species like rainbow trout, other salmonids, tilapia and catfish, successful rearing is generally possible at densities in which all fish of the rearing unit form a community. Such fish species thrive best in groups and may develop dominant and aggressive behaviour if there are too high or too low stocking densities, or if there is only one gender. Because of this, deciding on stocking densities on a fish farm is not a simple discussion to have, even though farmers usually prefer higher stocking densities as they assume this would automatically increase their production capacity.

However, based on research and experience, stocking density should be carefully considered and always supported by research and welfare guidelines.

How to Measure Stocking Density

To find the current stocking density of an already stocked growing system, one needs to have the following preliminary information:

1) The volume of water in the growing system,

2) The volume of the growing system, and

3) The number and weight of the fish stocked.

To calculate stocking density, the simple formula is:

$$\frac{Total\ number\ or\ weight/biomass\ of\ fish\ stocked}{Volume\ of\ water\ in\ the\ growing\ system}$$

= either Biomass/volume or number/ volume.

Using biomass is preferable because it captures the growth current state of the fish better than mere numbers. For instance, 10 fish weighing 500g each will occupy more space than 10 fish weighing 100 g each; thus, using numbers without regard for the growth stage can be misleading.

So, with this formula, it is assumed that in a pond with a total volume of 10,000 litres of water carrying 6,000 litres of water with 1,500 fish weighing 400g each,

Total biomass= 400g x 1500 = 600,000g (600kg)

Stocking density will be = 600,000/6,000 = 100g of fish per litre of water

Therefore, the stocking density of that growing system will be 100g of fish per litre (going by weight/biomass of fish) of water or ¼ fish per litre (going by number of fish). Before embarking on fish farming, the stocking density for the desired species (established by research and guidance) must be known and strictly adhered to. Furthermore, the feeding habits and natural behaviours of the species in question must be known and factored into the stocking density computations, as this will enhance productivity and welfare.

Recommended Stocking Densities

1. **Freshwater species**
    - Tilapia (*Oreochromis spp*.):
        - Ponds: 20-50 kg/m³. This density depends on aeration and water quality. Mechanical aeration can increase pond capacity. (Boyd, 1998)
        - Recirculating systems (RAS): Up to 100 kg/m³, with careful management of dissolved oxygen and organic loads (Timmons and Ebeling, 2010).
        - Impact on welfare: Above 100 kg/m³, the risk of stress and disease increases, with impacts on growth and survival (El-Sayed, 2006).

- Common carp (*Cyprinus carpio*):
  - Ponds: 10-20 kg/m³, suitable for semi-intensive systems where carp feed partially on natural sources (FAO, 2020).
  - Intensive ponds: 30-50 kg/m³, ensuring constant oxygenation (>4 mg/L) (Boyd and Tucker, 1998).
- Rainbow Trout (*Oncorhynchus mykiss*):
  - Raceways: 20-60 kg/m³, with a constant flow of water allowing for optimal oxygenation. (Conte, 2004)
  - RAS: Up to 80 kg/m³, but requires strict control of ammonia and nitrite. (Timmons et Ebeling, 2010)

2. Marine species

- Atlantic salmon (*Salmo salar*):
  - Floating cages: 10-20 kg/m³. Densities vary with depth and sea currents. Insufficient oxygenation can cause significant mortalities (Braithwaite *et al.*, 2006).
  - RAS: Up to 80 kg/m³. Recirculating systems are particularly suitable for reducing environmental impact and improving yields (Timmons and Ebeling, 2010).
- European sea bass (*Dicentrarchus labrax*) and gilthead sea bream (*Sparus aurata*):
  - Floating cages: 15-25 kg/m³, with attention to temperature variations and currents (FAO, 2020).
  - RAS: 40-70 kg/m³. These densities require careful management of the balance between oxygen and waste (Rakocy *et al.*, 2006).
- Shrimp (*Litopenaeus vannamei*):
  - Semi-intensive ponds: 5-10 ind./m², with additional nutritional support. (FAO, 2020)

    - Intensive systems (Biofloc): 300-500 ind./m³, where bioflocs act as an additional source of nutrition and improve water quality. (Avnimelech, 2012)

Q&A Session

- How do stocking densities affect oxygenation requirements in different systems (ponds, RAS, raceways)?
- What are the most reliable biological indicators for detecting density-related stress?
- Are there specific species that tolerate high densities better due to their social behaviour?
- How can RAS systems reduce the environmental impact of high densities?
- How does energy consumption vary between open systems (cages) and closed systems (RAS)?
- To what extent can artificial intelligence and automation optimise the management of aquaculture systems?
- Are there hybrid models that combine the advantages of several systems, such as aquaponics integrated into a RAS?

MODULE 5 – WATER QUALITY AND FISH WELFARE

Water quality is a key factor influencing the growth, health and welfare of fish in aquaculture. Each farming system (ponds, floating cages, raceways, recirculating systems) imposes specific requirements on water quality. An imbalance in physicochemical parameters can lead to stress, diseases and even high mortality.

Essential Water Quality Parameters

- **Dissolved Oxygen (DO):** Essential for fish respiration and microbial activity.
  - A minimum concentration of 5 mg/L is generally required, although some species tolerate lower levels.
  - DO levels can be optimised by mechanical aeration (diffusers, paddle wheels) and water flow management (especially in raceways and RAS) (Boyd and Tucker, 1998).
- **Temperature:** Depends on the specific needs of each species (e.g., trout: 10-18°C; tilapia: 24-30°C).
  - A large deviation can slow growth, induce stress and promote disease.
  - In closed systems such as RAS, heat exchangers can be used to stabilise the temperature (Timmons and Ebeling, 2010).
- **pH:** Most fish tolerate a pH between 6.5 and 8.5.
  - Too low a pH (<6) can cause osmotic stress, while too high a pH (>9) increases ammonia toxicity.
  - Adding calcium carbonate ($CaCO_3$) to ponds helps buffer pH variations (FAO, 2020).
- **Ammonia (NH_3/NH_4^+) and Nitrites (NO_2^-):** Un-ionised ammonia (NH_3) is toxic even at low concentrations (>0.05 mg/L).
  - Nitrites (>0.2 mg/L) affect the blood's ability to carry oxygen (brown blood disease).

- In RAS, biofiltration (biological filters) is essential to convert ammonia to nitrates (NO_3^-), which are less toxic (Boyd, 1998).

- **Hardness and Alkalinity:** Moderate hardness (50-150 mg/L $CaCO_3$) promotes good ionic balance for fish.
    - Sufficient alkalinity (>50 mg/L) is necessary to stabilise pH and provide carbonates to nitrifying bacteria (Boyd *et al.*, 2016).
- **Suspended Matter and Turbidity:** Excessive turbidity can impair fish respiration and limit light penetration (impacting photosynthesis in ponds).
    - The use of decanters and mechanical filters is recommended to reduce suspended matter (Timmons and Ebeling, 2010).
- **Salinity:** Essential for certain species such as sea bass or salmon, which require water adapted to their growth phase.
    - Poorly controlled salinity can cause significant osmotic stress (FAO, 2020).

Table 3 Water Quality Management in Aquaculture Systems

System	Water quality benefits	Disadvantages and challenges
Ponds	Natural balance of nutrients, natural biological regulation.	Risk of eutrophication, variations in temperature and oxygen.
Floating cages	Access to natural water, good water renewal.	Vulnerability to external pollution and uncontrollable environmental conditions.
Raceways	Good oxygenation due to constant flow.	Dependence on a clean and abundant water source and effluent management.
RAS (Recirculation)	Full control of parameters, low water consumption.	High costs, need for rigorous maintenance of biofilters and aeration systems.
Aquaponics	Waste reduction through integration with plant cultivation.	Complex management of nutrient balances for fish and plants.

Best Practices for Maintaining Optimal Water Quality

Water quality is one of the most crucial factors influencing fish health, and their entire existence depends on the water environment in which they live. This makes fish very sensitive to pollution and poor water quality issues. On the other hand, they will flourish and remain in good health in an optimal water environment that suits them. To ensure good water quality for optimal fish health and welfare, the following must be taken into consideration:

- Regular monitoring: Use of automatic sensors and analysers to measure oxygen, ammonia and pH.
- Efficient ventilation: Installation of aerators to maintain an optimal oxygen level, particularly in intensive systems.
- Biofiltration in RAS: Use of biological filters to transform ammonia into nitrates.
- Effluent management: Installation of sedimentation basins or mechanical filters to limit pollutant discharges.
- Feeding control: Avoid overfeeding, which promotes the accumulation of organic waste and toxic compounds.
- Thermal variation management: Use of heating or cooling equipment in closed systems.
- Infrastructure maintenance: Regular cleaning and disinfection to prevent the accumulation of harmful pathogens and biofilms.

Rigorous water quality management is essential to ensure optimal growth and limit stress and diseases of fish in aquaculture. Each system imposes specific challenges, requiring adapted strategies to maintain ideal conditions. Automation and technological innovation now offer advanced solutions for better control of water parameters, thus reducing environmental impacts and improving the sustainability of aquaculture.

Life Stages and Species-Specific Considerations

Water quality requirements vary for different species of fish and even for the different stages of their life cycles. The following table presents the general water quality parameters required for farmed Sea Bass, Sea Bream and Tilapia:

Table 4 Water quality parameters for sea bass, sea bream and tilapia

Species	Temp. (°C)	Dissolved Oxygen (mg/L)	pH	Salinity (%)	Total Ammonia mg/L)	References
Sea Bass (*Dicentrarchus labrax*)	15-25	> 5	7.5 - 8.5	30 – 40	< 0.02	FAO (2020), Espinosa *et al.* (2015)
Sea Bream (*Sparus aurata*)	18 - 28	> 5	7.5 - 8.5	30 - 40	< 0.02	FAO (2020), Tacon and Metian (2015)
Nile Tilapia (*Oreochromis niloticus*)	24 - 32	> 4	6.5 - 8.5	0 – 5	< 0.05	Boyd (1998), El-Sayed (2006)

Sea bass and sea bream welfare and water quality

The sea bass (*Dicentrarchus labrax*) and the gilthead sea bream (*Sparus aurata*) are two major species in Mediterranean aquaculture, particularly in Tunisia. Their well-being is closely linked to the quality of the water in which they live.

Table 5 Comparison between sea bass and sea bream welfare parameters

Parameter	Sea bass	Sea bream	Impact of welfare
Temperature (°C)	15-25	18-28	Temperature outside this range can cause stress, reduced growth and increased mortality (FAO, 2020).
Dissolved Oxygen (mg/L)	> 5	> 5	Too low a rate leads to hypoxia, stress and abnormal behaviours (Espinosa *et al.*, 2015).
pH	7.5 - 8.5	7.5 - 8.5	An inappropriate pH affects respiration and osmoregulation (Tacon and Metian, 2015).

Salinity (%)	30 - 40	30 - 40	Extreme variations can cause osmotic stress (FAO, 2020).
Total Ammonia (mg/L)	< 0.02	< 0.02	High concentrations of ammonia are toxic, causing gill lesions and stress (Boyd, 1998).
Turbidity	Moderate	Moderate	Excessively turbid water reduces visibility and impairs feeding behaviour (Huntingford *et al.*, 2006).

How to Measure and Correct Water Quality Parameters

Measuring water quality is essential for maintaining a healthy aquatic environment. Farmers can use various testing kits, electronic meters, or send samples to a water quality laboratory for more comprehensive analysis. For accurate measurements using test kits and meters, follow the instructions provided with the kit.

Table 6 How to solve some identified problems in water quality parameters

Parameter	Identified problem	Corrective solution
Temperature	Too high or too low	Adjust heating or ventilation, use heat exchangers
Dissolved Oxygen	Low level	Add aerators or inject pure oxygen
pH	Too acidic (< 7) or too basic (> 9)	Add baking soda (to raise pH) or vinegar/hydrochloric acid (to lower it)
Salinity	Too high or too low	Adjust with fresh water or seawater
Ammonia (NH_3)	High rate	Improve biological filtration, reduce feeding, and increase aeration
Nitrites (NO_2^-)	High rate	Partially change the water, and strengthen biological filtration
Nitrates (NO_3^-)	High rate	Increase water changes, promote aquatic plants or algae in the system
Turbidity	Cloudy water	Adjust power supply, improve mechanical filtration

Proactive and rigorous management of water quality helps ensure fish health and optimise the profitability of aquaculture farms.

Q&A Session

- What are the most important physicochemical parameters for water quality in aquaculture?
- What are the main water pollutants in aquaculture, and how can they be controlled?
- How to optimise water oxygenation in different aquaculture systems?
- What are the advantages and disadvantages of recirculation systems for water management?
- What are the effects of low dissolved oxygen on fish behaviour?
- How do aquaponics systems improve water quality management?

MODULE 6 – FEEDING AND FISH WELFARE

Feeding is a crucial factor for the growth, health and welfare of fish in aquaculture. Effective feeding management not only optimises zootechnical performance but also limits environmental and economic impacts.

- **Food choice**
  - **Species-specific formulation:** Nutritional requirements vary among species and growth stages. It is essential to provide a diet rich in specific proteins, lipids, carbohydrates, vitamins and minerals (NRC, 2011).
  - **Pellet size and texture:** Feed size should be appropriate for the fish's mouth to ensure efficient ingestion and reduce wastage (FAO, 2020).
  - **Use of quality ingredients:** Favour digestible protein sources to minimise nitrogen and phosphorus emissions (Tacon and Metian, 2008).
  - **Sustainable alternatives:** Incorporating plant or insect proteins to reduce reliance on fishmeal and fish oil (Henry *et al.*, 2015).
- **Ration management and feeding frequency**
  - **Respect for physiological needs:** Adapt the food ration according to the species, age, water temperature and breeding method (Kaushik and Troell, 2010).
  - **Feeding frequency:**
    - ✓ Fry: 4 to 6 meals per day
    - ✓ Juveniles: 2 to 4 meals per day
    - ✓ Adults: 1 to 2 meals per day
  - **Consumption control:** Observe eating behaviour to adjust rations and avoid overfeeding.
- **Adapted feeding techniques**
  - **Manual feeding:** Ideal for observing fish and adjusting rations according to their appetite.

- **Vending machines:** Ensure regular and controlled distribution, reducing stress and competition.
- **Demand feeding:** Allows fish to self-regulate by activating a distribution mechanism (Brett and Groves, 1979).

- **Impact of food on water quality**
  - **Reducing discharges:** Adjust the amount of feed distributed to avoid the accumulation of organic matter (Boyd and Tucker, 1998).
  - **Optimising feed conversion ratio (FCR):** A low FCR indicates better nutrient assimilation and reduced environmental impact (Timmons and Ebeling, 2010).
- **Adaptation to species-specific needs**
  - **Carnivorous species (e.g. sea bass, trout, salmon):** High requirement for animal protein (40-50%).
  - **Omnivorous species (e.g. tilapia, carp):** Mixed diet with a balance between animal and vegetable proteins.
  - **Herbivorous species (e.g. some carp, mullet):** Rich in plant fibre and protein (FAO, 2020).

Composition and Quality of Feed Ingredients

The quality of ingredients used in fish feed directly influences their growth, health and the environmental impact of aquaculture:

Table 7 Feed ingredients for fish

Category	Description
Proteins	**Animal origin:** Fishmeal and oil: rich in essential amino acids, expensive and limited availability. Fishing by-products: sustainable recovery of processing co-products. **Plant origin:** Soy, peas, algae: sustainable alternatives, possible antinutritional factors. **Proteins from insects**: High digestibility and good amino acid profile (Henry *et al.*, 2015).
	Omega-3 and Omega-6: Essential for growth and immune health.

Lipids	**Marine sources:** Fish oil: well assimilated but high environmental impact. **Plant-based alternatives:** Linseed oil, microalgae oil: rich in polyunsaturated fatty acids (Tacon & Metian, 2008).
Carbohydrates	**Energy sources:** Wheat, corn, potato starch. **Adaptation according to species**: Limited tolerance for carnivorous species.
Vitamins and minerals	Essential for growth, immunity and reproduction. **Supplementation:** Essential to avoid nutritional deficiencies (NRC, 2011).
Food additives	**Probiotics and enzymes:** Enhance digestion and nutrient assimilation. **Pigments:** E.g. astaxanthin: influences the colour of fish-like salmon. **Antioxidants and preservatives:** Ensure food stability and preservation (Merrifield *et al.*, 2010).

Q&A Session

- What are the specific nutritional needs of farmed species, and how are they integrated into feed formulation?
- How do alternative ingredients, such as plant or insect proteins, affect fish growth and health?
- How do meal frequency and distribution methods influence fish feeding behaviour and welfare?
- What are the key water quality parameters to monitor, and how does diet influence these parameters?

MODULE 7 – FISH WELFARE DURING HANDLING AND TRANSPORTATION

Fish handling and transport are critical steps that can lead to stress, injury and loss if not properly controlled. A careful approach, based on appropriate protocols, can reduce negative impacts and ensure the viability of fish after transport (Ashley, 2007).

Stress Factors during Handling

Fish are particularly sensitive to interactions with their environment and physical disturbances. Major stressors include:

- **Rough capture and handling:** Excessive or improper handling can cause bruising, skin damage, and loss of protective mucus, increasing the risk of infections (Harmon, 2009).
- **Time out of water:** Fish do not have structures to breathe air, so prolonged out-of-water exposure can lead to asphyxiation and physiological imbalances (Ross and Ross, 2008).
- **Water temperature:** Sudden temperature changes between tanks and handling areas can cause thermal shock and affect fish metabolism (Ashley, 2007).
- **Dry handling:** Fish have protective mucus that is essential to their health. Handling with dry hands or on rough surfaces can damage this natural barrier (Harmon, 2009).

Transportation and Fish Welfare

Fish transport is a critical step in aquaculture, influencing their welfare, health and survival. Poorly managed transport can result in acute stress, injury, reduced water quality and, in extreme cases, high mortality (Harmon, 2009). It is therefore essential to apply rigorous protocols to minimise these negative effects.

- **Stress Factors During Transportation:** Transport exposes fish to several sources of stress, which can impact their physiology and behaviour:

- Excessive handling: Capturing, sorting, and containerisation can cause injury and loss of protective mucus, increasing susceptibility to infections (Ross and Ross, 2008).
- High stocking density: Overcrowding in transport containers leads to competition for oxygen, accumulation of metabolic wastes, and increased stress (Harmon, 2009).
- Variations in water parameters: Sudden changes in temperature, oxygen, pH, or salinity can cause physiological shock (Ashley, 2007).
- Transport duration: The longer the journey, the greater the risk of stress, exhaustion, and mortality (Harmon, 2009).
- Vibration and movement: The jolts and noise of transport disrupt the balance of fish and increase their stress levels (Ross and Ross, 2008).

Best Practices for Safe and Humane Transportation

1. **Preparation before transport**
   - **Pre-fasting:** It is recommended not to feed fish for 12 to 48 hours before transport to limit waste production and improve water quality (Harmon, 2009).
   - **Gradual adaptation:** Slowly adjust container water parameters to avoid thermal and osmotic shock (Ashley, 2007).
   - **Fish selection:** Check the health of individuals and avoid transporting sick or injured fish (Ross and Ross, 2008).
2. **Optimal transport conditions**
   - **Proper Stocking Density:** Stocking density should be adjusted based on species, temperature, and duration of transport to minimise stress and asphyxiation (Harmon, 2009).
   - **Oxygenation:** Maintain dissolved oxygen concentrations above 5 mg/L using oxygen diffusers or air injection (Ashley, 2007).

- **Temperature Control:** Use coolers or cooling systems to maintain a stable temperature appropriate for the species being transported (Ross and Ross, 2008).
- **Water Filtration and Treatment:** Add agents such as salt (to reduce osmotic stress) or anti-ammonia to improve water quality (Harmon, 2009).
- **Transport equipment:** Use suitable containers with rounded edges to avoid injury, and opaque containers to limit exposure to light and reduce stress (Ross and Ross, 2008)

The transport of sea bass (*Dicentrarchus labrax*) and sea bream (*Sparus aurata*) raised in Tunisia follows strict protocols to minimise stress and ensure their well-being.

3. Means of Transport

- **Transport in on-board tanks:** Fish are transferred into trucks equipped with tanks or reservoirs specially designed to maintain optimum water quality. These tanks are equipped with oxygenation and temperature control systems to reduce stress on the fish during transport. (Figures 16 and 17).
- **Transport in plastic bags (for juveniles):** Mainly used for juveniles or over short distances, this method involves placing the fish in bags filled with water and pure oxygen, then sealing them to ensure a controlled atmosphere.

Figure 16 Examples of trucks with oxygenated tanks to transport fish

Figure 17 Tanks to transport fish

4. **Equipment Used**
   - **Oxygenation Systems:** Oxygen diffusers are installed in tanks to maintain adequate levels of dissolved oxygen, essential for the survival and well-being of fish (fao.org).
   - **Temperature controllers:** Heating or cooling devices help maintain water at an optimal temperature, preventing thermal shock (linde.ch).
   - **Filtration systems:** To remove waste and maintain proper water quality, mechanical and biological filters are integrated into the transport tanks.
   - **Monitoring Equipment:** Sensors and monitors continuously measure water parameters such as temperature, pH and oxygen levels, allowing for real-time adjustments (aquaculturefrance.com).

Q&A Session

- What are the main stressors for fish during transport, and how can they be mitigated?
- How does loading density influence fish welfare during transport?
- What are the impacts of water temperature variations on fish health during transport, and what measures can be taken to control them?
- What specific training should operators undergo to ensure fish welfare during transport operations?

MODULE 8 – SLAUGHTERING AND FISH WELFARE

Overview of Humane Fish Slaughter

Humane slaughter of fish aims to minimise their suffering during killing by ensuring rapid loss of consciousness and avoiding unnecessary pain. However, current practices in the fish farming industry often fall far short of this ideal. In Europe, while regulations require terrestrial animals to be stunned before slaughter, fish do not always benefit from these protections. As a result, cruel slaughter methods, such as open-air asphyxiation, remain legal in some contexts, despite the suffering they cause (www.l214.com).

WOAH's *Aquatic Animal Health Code* provides guidelines on fish welfare during stunning and slaughter, which member states are expected to incorporate into their national regulations. In response, several relatively humane slaughter methods have been developed. Although many of these methods are not yet perfect, they represent progress compared with older, clearly inhumane practices that are now widely rejected or banned. Generally accepted methods include those using electrical stunning, as they are rapid and have been shown to cause minimal physical and physiological impact on the fish.

Furthermore, it is highly recommended that stunning and slaughter of fish must be conducted by staff who have the technical capacity, training, and knowledge to utilise slaughter equipment, can recognise when effective stunning has taken place, and know how and when to re-stun, if necessary. They should receive periodic retraining, upskilling, and evaluation of their stunning and slaughter methods, and keep records of these activities on the farm. This is especially important because fish slaughter equipment and methods are still evolving as fish welfare and industry professionals continue to make efforts to ensure a seamless and painless slaughter process.

Benefits of Humane Slaughter of Fish

It is important to note that unstunned slaughter is generally associated with increased suffering for animals. Fish, like other animals, experience pain and stress. Unstunned slaughter methods, such as open-air asphyxiation or ice bath immersion, can result in prolonged agony and significant distress for fish.

In contrast, pre-slaughter stunning aims to render the animal unconscious, so that it does not experience the pain and distress associated with incision and exsanguination during slaughter.

Therefore, slaughter with pre-stunning is generally recommended to *minimise* animal suffering and improve the quality of the final product.

Carrying out humane slaughter of fish offers several benefits for the fish, the farmer, and consumers. These are elucidated below:

- Humane slaughter methods improve meat quality and reduce the risk of spoilage (Fish Count, 2019). It reduces the appearance of soft flesh, gaping, bruising, and scale loss, and improves shelf life when compared to traditional, less humane slaughter methods (Holmyard, 2017). For example, fish slaughtered with more humane methods often have firmer, translucent fillets with brighter colours, and the onset and severity of rigour mortis are delayed when compared to conventional, less humane slaughter methods (Humane Slaughter Association, 2019).
- Reducing stress at slaughter through humane slaughter methods is also likely to improve eating quality and taste for the consumer (Fish Count, 2019).
- Implementing humane slaughter processes increases the ethical value of the fish product, which can potentially add economic value. Ethical consumers are often willing to pay extra for fish that is produced and slaughtered more humanely (Fish Count, 2019).
- Practising humane methods of slaughter improves compliance with existing local and global food processing and safety standards, which invariably improve the market value of the product.

Pre-slaughter Welfare Considerations

Ensuring fish welfare prior to slaughter is essential to minimise suffering and ensure optimum quality of the final product. Key considerations include:

1. Pre-slaughter fasting

Before slaughter, it is recommended to fast fish for a period of time to empty their digestive system. This practice reduces the risk of contamination of the flesh during gutting and reduces stress related to handling. However, the fasting period must be carefully adjusted to prevent any decline in fish quality. For example, one study indicates that a 24-hour fast improves product quality compared with longer fasting periods (https://www.agrociwf.fr/).

2. Handling and transport

Handling and transportation operations can be a significant source of stress for fish. It is therefore crucial to limit handling and ensure that fish are loaded and unloaded without exposure to air, while maintaining high oxygen levels in the water and appropriate stocking density (EFSA, 2017).

3. Stunning Methods

Stunning before slaughter is essential to avoid unnecessary pain to the fish. Stunning methods must cause immediate and irreversible loss of consciousness. If this is not the case, the fish must be slaughtered before they regain consciousness (Escudero, 2018).

4. Slaughter Methods

After stunning, slaughter must be carried out quickly and efficiently to avoid prolonged suffering. Methods such as *ikejime*, a traditional Japanese technique, involve puncturing the fish's brain, which minimises stress and preserves the quality of the flesh (lemonde.fr).

5. Staff training

Staff involved in fish slaughter must be trained in good handling and slaughter practices to ensure animal welfare and the quality of the final product.

By implementing these measures, it is possible to significantly reduce the stress and suffering of fish before slaughter, while ensuring a better quality of products intended for consumption.

Common Fish Slaughter Methods

Fish slaughter is a crucial step in aquaculture and fisheries, influencing both animal welfare and the quality of the final product. The methods used vary in terms of efficiency and respect for animal welfare. Here is an overview of the main techniques used:

1. Open-air asphyxiation

This traditional method involves removing fish from the water and allowing them to die by asphyxiation. It is commonly used in many fish farms. However, this practice can cause prolonged suffering, as fish can take more than an hour to die while struggling to breathe (https://www.agrociwf.fr/).

2. Ice bath immersion

Fish are placed alive in a mixture of water and ice, causing progressive hypothermia until death. Although this method is commonly used, it is criticised for the stress and pain it can cause the fish before they lose consciousness (https://welfarm.fr/).

3. Electrical stunning

This technique exposes fish to an electric current of sufficient intensity and frequency to cause immediate loss of consciousness. It is considered more humane and reduces animal suffering while improving the quality of the final product (https://www.publications.gov.on.ca/)

4. Mechanical Percussion

The use of a captive bolt percussion gun, with or without penetration of the rod, causes a massive and instantaneous interruption of brain function, rendering the animal unconscious. This method is effective in ensuring rapid loss of consciousness (https://inspection.canada.ca/).

5. *Ikejime* Method

Originating in Japan, *ikejime* is a traditional technique that involves puncturing the fish's brain, followed by the destruction of the spinal cord, to minimise stress and preserve the quality of the flesh. This method is increasingly being adopted outside Japan for its benefits in terms of animal welfare and product quality (Poli *et al.*, 2005).

It is essential to note that certain methods, such as the use of carbon dioxide (CO_2) for tourism, are recommended as they are suitable for fish and do not contravene the standards of the World Health Organisation (WHO).

Overview of the Slaughter Process in Tunisia

1. Regulatory framework and welfare standards

In Tunisia, animal welfare regulations in aquaculture remain under development, but some international guidelines, such as those of the World Organisation for Animal Health (WOAH), influence local practices.

- Slaughter mainly follows health standards to ensure the quality of the final product intended for local consumption and export.
- Veterinary controls ensure compliance with hygiene and food safety conditions.

2. Slaughter methods used

Practices vary depending on the size of the farms and the equipment available:

Table 8 Slaughter methods

Method	Advantages	Disadvantages
Traditional slaughtering Used in small farms and local markets: **Asphyxiation in air or on ice:** Fish are placed on ice or left in the open air until movement stops. **Immediate decapitation:** Faster method but rarely *practised* due to the difficulty of handling.	Ease of execution and low cost.	Slow method, stressful for the animal and affects the quality of the flesh.

Modern methods of slaughtering In large fish farms and export units, techniques that are more respectful of animal welfare and product quality are being adopted:		
Electrical stunning: A rapid method that causes immediate loss of consciousness before slaughter.	Stress reduction, preservation of flesh texture and colour.	Requires specific equipment.
Slaughter by immersion in ice water (Most common method): Fish are placed in a mixture of water and ice, causing a gradual loss of consciousness before the cessation of vital functions.	Common practice in Tunisia, this approach is economically viable and acceptable for exports.	Prolonged loss of consciousness can cause stress.
***Ikejime* method (practised to a limited extent):** Japanese technique of immediately perforating the brain for rapid death and better preservation of the flesh.	Excellent fish quality, optimal preservation of taste and texture.	Little known in Tunisia and requires specific know-how.

3. Challenges and perspectives

- Lack of strict regulation: The lack of clear national guidelines on animal welfare in aquaculture can limit the application of best practices.
- Modernisation of infrastructure: Large farms are looking to invest in more efficient slaughtering equipment to meet the demands of international markets.
- Raising awareness among producers: Better training of aquaculture farmers on animal welfare-friendly techniques could improve current practices.

Fish slaughtering in Tunisia still relies mainly on traditional methods, although large companies are gradually adopting more advanced techniques to meet international standards. Improving infrastructure and adopting new technologies could, in the future, strengthen humane slaughter practices and optimise the quality of products from the Tunisian aquaculture sector.

Q& Session

- What are the most commonly used slaughter methods in Tunisia for the main farmed species, such as sea bream and sea bass?
- To what extent do international guidelines, such as those of the World Organisation for Animal Health (OIE), influence Tunisian fish slaughter practices?
- How do slaughter practices influence local and international consumers' perceptions of the sustainability and quality of Tunisian aquaculture products?

MODULE 9 – ENVIRONMENTAL ENRICHMENT AND FISH WELFARE

What is Environmental Enrichment?

Environmental Enrichment (EE) involves enhancing an animal's living environment to promote species-specific behaviours, mental stimulation, and overall well-being. In the context of fish, it refers to creating conditions that mimic their natural habitats and encourage natural behaviours. It can include adding structures or modifying rearing units to create a more natural or complex environment that resembles the fish's natural habitat. It may also include any intentional augmentation of complexity in the surroundings of the animal, such as structures made from plants and pebbles, music, unusual foods, and the introduction of various fish species. Furthermore, it may include mimicking colours and introducing varied conditions, such as dark hiding spots and cooler water areas, for them to choose from (Leone and Estévez, 2008; Näslund and Johnsson, 2014). This is particularly relevant in captive settings such as aquaculture farms and public aquariums (Zhang *et al.,* 2020).

- ⇨ Environmental enrichment in aquaculture aims to improve fish welfare by recreating elements of their natural habitat, thereby promoting the expression of innate behaviours and reducing stress. This approach, widely studied in terrestrial animals, is also being increasingly applied to aquatic species.

The challenge is determining the type and quantity of environmental enrichment that fish prefer, and this can be aided by understanding their sensory abilities. To get started, we must ensure that each potentially enriching material is pertinent to the biology and preferences of the species. For instance, some fish may prefer hiding, while others may prefer swimming against the current (Zhang *et al.,* 2020).

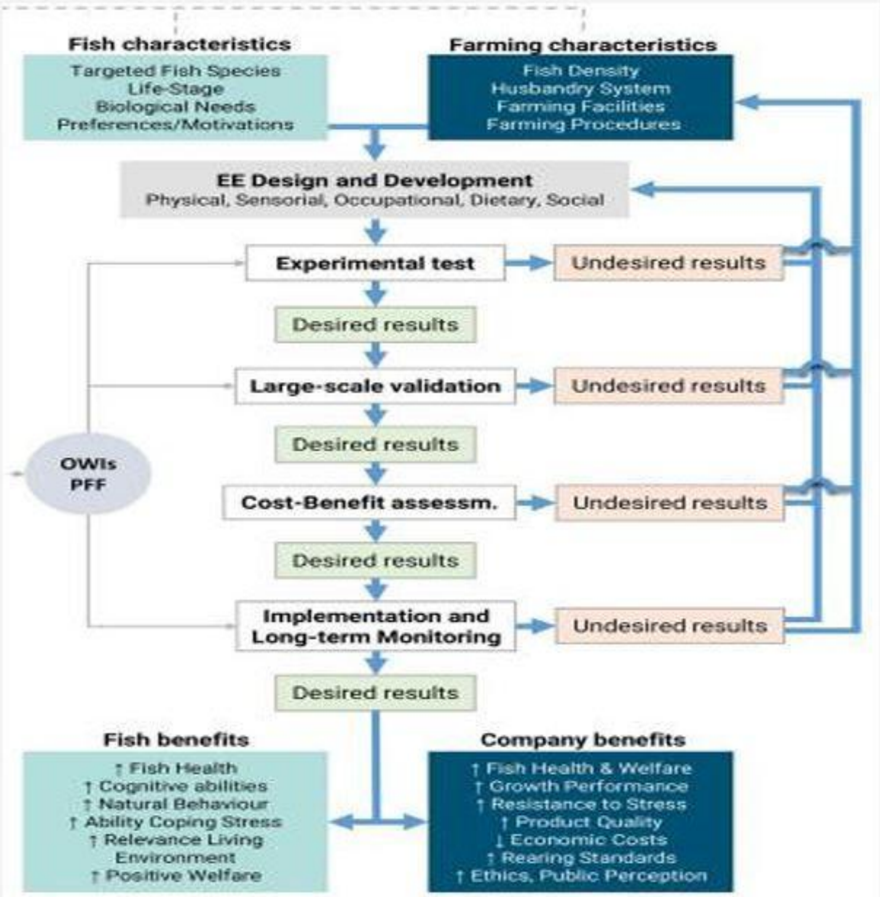

Figure 18 Schematic for the decision-making process in Environmental Enrichment; OWIs: Operational Welfare Indicators; PFF: Precision Fish Farming (Source: Arechavala-Lopez et al., 2021)

Types of Environmental Enrichment

1. **Physical Enrichment:** Introducing structures such as hiding places, varied substrates, or floating objects to stimulate exploration and provide refuge.
2. **Sensory Enrichment:** Using visual, auditory, or olfactory stimuli to engage fish's senses and encourage natural behaviours.
3. **Dietary Enrichment:** Diversifying food sources or feeding methods to stimulate foraging and physical activity.
4. **Social Enrichment:** Maintaining species-appropriate social groups to promote positive interactions and reduce aggression.

Benefits of Environmental Enrichment

Environmental enrichment in aquaculture has several benefits for fish welfare:

- Reduced Stress and Aggression: An enriched environment provides varied stimulation that reduces stress and aggressive behaviour in fish (https://www.agrociwf.fr/).
- Improved physical and mental health: Appropriate enrichments allow fish to express natural behaviours, contributing to their overall well-being (https://www.agrociwf.fr/).
- Increased resilience and adaptability: Stimulating environments promote the development of adaptive capacities in fish, making them more resilient to environmental changes or stresses (https://chaire-bea.vetagro-sup.fr/).
- Improved Water Quality: An enriched environment can contribute to improved water quality, which is essential for fish health (https://www.agrociwf.fr/).

By incorporating appropriate enrichments, aquaculture producers can improve fish welfare, which can also lead to increased productivity and reduced health problems within the flock.

Species Recommendations for Environmental Enrichment

Fish responses to environmental enrichment vary widely and can be positive—such as improved cognitive performance and behaviour, reduced stress levels, and better physical condition (Arechavala-Lopez et al., 2020; Batzina and Karakatsouli, 2012; Bosakowski and Wagner, 1995; Soares et al., 2011)—or negative, including increased aggression and higher stress levels (Barreto et al., 2011; Sabri et al., 2012; Woodward et al., 2019). These responses differ among species, types of enrichment, life stages, and group sizes. Hence, it is essential to monitor behavioural, physical, and physiological welfare indicators when developing a new environmental enrichment protocol.

Gilthead sea bream

Gilthead sea bream (*Sparus aurata*) commonly swim between the surface and 30 m depth. They interact with the substrate to rest and hunt for food (Abecasis and Erzini, 2008; Jobling and Peruzzi, 2010); therefore, the addition of substrate could provide welfare benefits to sea bream in aquaculture.

Structural enrichment

Gilthead sea bream (juveniles, 20.3 ± 0.22 g) reared for 75 days with blue substrate (glass gravel 6-12 mm in size) displayed less aggressive behaviour and had lower basal cortisol levels than sea bream reared without substrate (Batzina, Kalogiannis, *et al.,* 2014). Blue or red-brown gravel has been found to lower aggression; however, this effect was not observed in fish exposed to green gravel (68.3 ± 0.50 g, exposed for 84 days, with glass gravel 6-12 mm in size) (Batzina and Karakatsouli, 2012).

In a substrate-colour preference test (glass gravel 6-12 mm in size), 2-year-old gilthead seabream (84.8 ± 1.9 g) preferred blue substrate over red-brown, green, or no substrate, but juvenile sea bream (21.9 ± 0.5 g) showed a preference for red-brown over green substrate but did not show a preference when offered the choice between blue vs. green or blue vs. red-brown substrate (Batzina, Sotirakoglou, *et al.,* 2014). Substrate can benefit the welfare of sea bream, but the reason why a difference in colour preference was observed between the two age groups is unclear, and further research is needed to ensure optimal enrichment can be provided to gilthead sea bream at each life stage.

When comparing the effect of exposing juvenile sea bream (20.2 ± 0.26 g) to blue substrate (glass gravel 6-12 mm in size, structural enrichment) vs. a photo of blue substrate (sensorial enrichment simulating structural enrichment) for 74 days, the fish exposed to substrate were less aggressive and were observed manipulating the substrate (Batzina and Karakatsouli, 2014). These studies show the importance of substrate and substrate colour on the welfare of gilthead sea bream. The observations made by Batzina and Karakatsouli (2014, 2012) and Batzina *et al.* (2014b) that sea bream manipulated the substrate are important. Gilthead sea bream interact with the substrate in the wild for either environmental exploration (e.g. foraging) or predator protection (Abecasis and Erzini, 2008; Jobling and Peruzzi, 2010); therefore, providing captive gilthead sea bream with manipulable substrate could allow them to perform ethologically important behaviours. All the studies reviewed on the benefits of gravel substrate for the welfare of sea bream were carried out at a laboratory scale in small tanks. Research is needed to determine the effect of gravel on the welfare of gilthead sea bream in large-scale aquaculture systems.

Sensory enrichment

In a study researching sensory enrichment similar to that discussed above in rainbow trout, Papoutsoglou *et al.* (2008) exposed juvenile gilthead sea bream (1.51 ± 0.01 g) to Mozart for 2 or 4h per day using a hydrophone in either a high or low lighting environment for 117 days. Fish exposed to Mozart for 4 h per day with high lighting had lower levels of brain neurotransmitters, indicating reduced stress. Music has the potential to reduce stress in gilthead sea bream reared on land in hatcheries, ponds, or raceways; however, further research is needed. It is important to note that the points raised by the authors in the rainbow trout study about the possible effect of environmental and life-stage variability on the benefits of musical enrichment must also be considered in gilthead sea bream and all other fish species exposed to this type of enrichment (Papoutsoglou *et al.*, 2013).

European sea bass

Enriched environments did not affect the growth performance of seabass at the group level. Different studies indicate that the effects on growth can be positive, negative, or have no effect. In agreement with this study, Arechavala-Lopez *et al.* (2019) found that vertical enrichment did not affect the growth of juvenile gilthead seabream. Similarly, in other aquaculture species reared under enrichment conditions, the growth was no different from the control (Brockmark *et al.,* 2010; Roberts *et al.,* 2011; Ren *et al.,* 2019). However, positive effects of enriched environments have also been reported. For instance, the addition of a colour substrate to reared seabream enhanced growth performance and even reduced aggressiveness (Batzina and Karakatsouli, 2012, 2014; Batzina *et al.,* 2014a, b, c, d).

Rearing techniques for European sea bass are often similar to those of gilthead sea bream; however, further research is needed to determine the species-specific environmental enrichment that best improves the welfare of European sea bass in aquaculture.

Tilapia fish

Environmental enrichment strategies for Tilapia fish species have been studied to improve their behaviour and welfare in captivity. Studies have shown that structural environmental enrichment, such as the use of plant-fibre ropes or physical structures, can enhance cognition, exploratory behaviour, and brain physiological functions in Tilapia fish (Torrezani *et al.,* 2013). Enriched environments have been shown to reduce aggression and increase hierarchical behaviour in Tilapia fish (Arechavala-Lopez *et al.,* 2020). As adapted from the Aquatic Life Institute (ALI), key recommendations for environmental enrichment of Tilapia fish have been elucidated in Table 9 below.

Table 9 Environmental enrichment recommendations for the tilapia fish species

Nile tilapia (*Oreochromis niloticus*)		
Enrichment Category	**Juvenile**	**Adult**
Enclosure Coloration	Not enough information is available at this time. Therefore, we default to the species' "natural" conditions at this stage.	Maia and Volpato (2016) demonstrated that it takes at least 10 days of testing to determine the colour preference of Nile tilapia, and that green and blue are the most preferred colours by the species.

Substrate Provision	Enrichment with e.g. river pebbles and plastic kelp models probably increases the value for juveniles, but this may cause more intense fights to establish territories (FishEthoBase). Must be closely monitored.	Males tend to choose sand as their nesting substrate over other substrates, such as stones. Individuals presented equal frequency of total attacks, whether they were being kept with or without substrates, but fewer highly intense attacks were observed in animals kept with the substrate. For the most natural solution, provide sand and mud; alternatively, provide gravel. Bamboo poles also increase growth (FishEthoBase).
Lighting	Increased light intensity (280-1390 lx) reduces aggressive interactions between pairs of juvenile males. Natural photoperiod is 9-15 hours. Provide access to natural (or at least simulated) photoperiod and daylight. (FishEthoBase)	Blue light reduces stress by preventing the confinement-induced cortisol response (Volpato and Barreto, 2001). Natural photoperiod is 9-15 hours. Provide access to natural (or at least simulated) photoperiod and daylight. Avoid 1,400 lux, as it increases aggression compared to 280 lux. (FishEthoBase)
Water Augmentation	Depth: Provide at least 2-6 m, ideally up to 20 m, bearing in mind the planned stocking density. Individuals should be able to choose swimming depths according to life stage and status. (FishEthoBase)	Depth: Provide at least 2-6 m, ideally up to 20 m, bearing in mind the planned stocking density. Individuals should be able to choose swimming depths according to life stage and status. (FishEthoBase)
Structures	An enriched environment increases resource value, which in turn prompts more intense fights (FishEthoBase)	Fish cultured in environments enriched with artificial water hyacinth and shelter presented higher latency to trigger confrontations, and the confrontations were less intense in the section with enrichment items (Neto and Giaquinto, 2020).
Shelter	An enriched environment increases resource value, which in turn prompts more intense fights (FishEthoBase)	For the most natural solution, provide roots or submerged branches, bushes, or trees; alternatively, provide artificial shelters inside the system (e.g. artificial reef) (FishEthoBase)
Feeding System	Ensure that you provide sufficient feed from approximately 4-8 days after hatching. Self-feeders could	Tryptophan-supplemented food was found to reduce confrontations (Neto & Giaquinto, 2020). Install a self-feeder and make sure all Nile tilapia adapt to it.

	prevent stressful food competition (FishEthoBase)	(FishEthoBase) Provide sand, mud, and bamboo poles so that individuals may search for food. (FishEthoBase)

In conclusion, environmental enrichment is a powerful tool for enhancing fish welfare by providing opportunities for species-specific behaviours, mental stimulation, and improved overall health. Recognising the importance of environmental enrichment in captive settings can contribute to the ethical treatment of fish and the sustainability of aquaculture practices. Regular research and collaboration between scientists, aquaculturists, and conservationists will continue to advance our understanding of effective enrichment strategies.

Q&A Session

- What are the potential impacts of environmental enrichment on water quality in aquaculture systems?
- How can environmental enrichment contribute to the sustainability of aquaculture practices?
- Are there differences in environmental enrichment needs between farmed fish species?
- How does environmental enrichment influence the health and growth of farmed fish?
- What are the main objectives of environmental enrichment in aquaculture?

MODULE 10 – FISH HEALTH AND WELFARE

Animal Health and Welfare

Animal welfare is defined as the state of the animal, the treatment it receives from animal care and husbandry, and the humane treatment it experiences, as well as how an animal copes with the conditions in which it lives (Animal Welfare Institute, 2018). Animal Health can be defined as the absence of disease and the normal functioning of an organism, as well as normal behaviour (Ducrot *et al.*, 2011). From the above definitions, it is evident that the concepts of animal 'health' and 'welfare' are distinct yet closely linked. For example, an animal in a good state of welfare is considered healthy, comfortable, well-nourished, safe, able to express its innate behaviour, and is not suffering from unpleasant states such as pain, fear, and distress.

The main difference is that animal health primarily focuses on the occurrence, impact and treatment of diseases, infections and sub-optimal health conditions, while welfare incorporates the sentience and mental complexity of animals which includes their ability to feel emotions, have needs, be conscious and their ability to adapt to domestication without negatively impacting their freedom of expression of natural behaviors (Nicks and Vadenheede, 2014). Although varying in their approaches to well-being, they mutually impact each other and are both integral to the overall optimal well-being and livelihood of animals. Good animal welfare, especially for farmed animals, encompasses disease prevention, appropriate shelter, management, nutrition, humane handling and humane slaughter (Animal Welfare Institute, 2018). Therefore, the idea of welfare remains an important element in addition to traditional animal health concerns (Nicks and Vadenheede, 2014).

Biosecurity for Fish Health and Welfare

Biosecurity refers to a set of measures aimed at preventing the introduction, spread, and impact of infectious diseases in aquaculture production systems

(FAO, 2020). It is essential to ensure fish health and welfare, optimise productivity and reduce economic losses from disease outbreaks (WOAH, 2021).

According to Yanong and Erlacher-Reid (2012), the major goals of biosecurity are:

- Effective animal management through acquiring healthy fish stocks and optimising their health and immunity through good husbandry.
- Management of pathogens by preventing, reducing, or eliminating pathogens.
- Management of people by educating, training, and managing the movement of staff and visitors

Farmed fish are exposed to a variety of pathogens, including:

- Viruses (e.g. infectious haematopoietic necrosis virus, nodavirus) (FAO, 2020).
- Bacteria (e.g. *Aeromonas hydrophila*, *Vibrio spp*., *Flavobacterium columnare*) (Subasinghe *et al.*, 2019).
- Parasites (e.g. *Ichthyophthirius multifiliis, Gyrodactylus* spp.) (WOAH, 2021).
- Fungi (e.g. *Saprolegnia spp*.) (FAO, 2020).

The absence of strict control can lead to epidemic outbreaks, impacting production and threatening the biodiversity of natural environments (WOAH, 2021).

Benefits of Biosecurity on Fish Farms

Implementing biosecurity measures in fish farms has several major benefits, both for fish health and for the economic and environmental sustainability of aquaculture operations.

1. Disease prevention and economic loss reduction

Reducing disease outbreaks by preventing the introduction and spread of pathogens (FAO, 2020).

Reducing production losses caused by viral, bacterial and parasitic infections (WOAH, 2021).

Reducing costs related to veterinary treatments and antibiotics (Subasinghe *et al.,* 2019).

2. Improved fish health and welfare

Maintaining optimal stocking density to reduce stress and aggressive behaviour (FAO, 2020).

Improved water quality, ensuring a favourable environment for fish growth and development (WOAH, 2021).

Reduced medical interventions, avoiding the accumulation of drug residues in fish and the environment (Subasinghe *et al.,* 2019).

3. Increased productivity and profitability

Improved fish survival rates, leading to more stable and predictable production (FAO, 2020).

Optimisation of resources (water, feed, treatments) through rigorous control of farming conditions (WOAH, 2021).

Access to better markets and certification for fish products from systems that meet health standards (Subasinghe *et al.,* 2019).

4. Reducing environmental impact

Less discharge of chemical and biological contaminants into aquatic ecosystems (FAO, 2020).

Limiting the transmission of diseases to wild fish populations (WOAH, 2021).

Promoting sustainable practices, reducing reliance on antibiotics and other chemicals (Subasinghe *et al.,* 2019).

5. Compliance with regulations and access to international markets

Compliance with international health standards (e.g. WOAH, FAO) facilitating the export of products (WOAH, 2021).

Improved traceability and quality guarantees for consumers and investors (FAO, 2020).

Strengthening the reputation of fish farms, promoting their competitiveness in the global market (Subasinghe *et al.,* 2019).

Common Biosecurity Measures and Practices

The application of biosecurity in fish farming is based on a set of preventive measures aimed at reducing health risks, improving productivity and minimising environmental impact.

Bera *et al. (2018) and Ernst et al.* (2017) provide a comprehensive list of good biosecurity measures and practices that fish farmers should adopt. These include the following:

❖ Providing a clean, pathogen-free water source at all times for land-based fish farms.

❖ Restricting the movement of fish from one farm to another, especially from those of poorer health.

❖ Limiting visits to the fish farm or access to a farm site, i.e. by setting up gates and fences.

❖ Fixing clear signs to direct traffic within and outside the farm where necessary.

❖ Establishing and implementing strict sanitary measures, such as defining designated sanitary areas, cleaning and disinfecting for individuals entering the farm, using protective and disinfected clothing, implementing foot dips, and promoting hand hygiene.

❖ Restricting the movement of tools and culture organisms.

❖ Fish stock health should be maintained by keeping stock stress to a minimum level and maintaining optimum water quality.

❖ Minimise the pest and disease risk associated with stock movements onto, within and off your farm by maintaining appropriate quarantine procedures during stock movement.

❖ Minimise the risks of pests and disease entry associated with incoming water through proper treatment.

❖ Preventing the entry and spread of pests and diseases by assessing all equipment, vessels and vehicles entering the farm through proper biosecurity procedures like disinfection of equipment, controlled use, etc.

❖ Records should be kept of the workers and visitors, and all the workers should be trained on biosecurity standards.

❖ Food-borne disease organisms can be minimised by proper handling and storage.

❖ Implementing pest control management by controlling or eradicating predators, wildlife, scavengers, and other organisms from farm areas.

❖ Wastewater and solid waste should be treated appropriately before disposal.

❖ Maintain records for all aspects of the biosecurity plan (staff training, workers and visitors' log, inspection, and maintenance of farm infrastructure).

❖ Regular monitoring, surveillance and audit of the biosecurity measures should be implemented throughout the farm.

❖ Development and implementation of an appropriate biosecurity management plan (Bera *et al.,* 2018; Ernst *et al.,* 2017).

Fish Diseases and Impacts

Disease outbreaks pose a significant threat in aquaculture, resulting in substantial economic losses for farms due to increased mortality, decreased growth and productivity, and higher production costs. Due to its catastrophic impacts on aquaculture, FAO (2020) regarded it as one of the major obstacles to the growth and development of sustainable aquaculture. The major barriers to the effective prevention and control of diseases in fish farms include inadequate aquaculture disease management training, the unavailability of effective drugs within the reach of farmers, the high cost of quality feeds, the high cost of drugs and treatment, and poor financial support. These indicate the need for fish farmers and managers to be well-trained in aquaculture disease management, to reduce the occurrence of disease outbreaks, and to increase their farms' economic performance.

Numerous infectious diseases are significant to global aquaculture, and they are often caused by viruses, bacteria, parasites, fungi, or pests (Cascarano *et al.*, 2021). They have the capacity to spread through the movement of infected host species, have devastating effects on aquaculture productivity, and pose greater challenges for aquaculture development (Subasinghe *et al.,* 2009).

Fish diseases undermine sustainable development goals, particularly in developing nations, by reducing income earnings, leading to job losses, jeopardising food availability, and posing a threat to nutrition and food security (World Bank, 2014). Because aquaculture in developing nations is typically small-scale and rural, the vast majority of infections remain undetected, untreated, and unreported, placing a significant burden on populations struggling to overcome poverty (Mukaila *et al.,* 2023).

Diseases in fish and other animals can be caused by infectious organisms, such as bacteria, viruses, fungi, parasites, and protozoa, or may have miscellaneous non-infectious origins.

Table 10 Common bacterial diseases of farmed fish

Disease	**Pathogen**	**Affected Species**	**Symptoms**	**Transmission**	**Treatment and Prevention**	**Reference**
Vibrioses	*Vibrio anguillarum*, *Vibrio vulnificus*, *Vibrio salmonicida*	Salmon, trout, sea bass, sea bream	Septicemia, skin ulcers, exophthalmos (bulging	Contaminated water, contact with infected fish.	Antibiotics, vaccination, and improved farming conditions.	Toranzo *et al.*, 2005

			eyes), haemorrhages.			
Furunculosis	*Aeromonas salmonicida*	Salmon, trout, catfish	Boils (abscesses under the skin), lethargy, and internal bleeding.	Direct contact with sick fish or contaminated water.	Vaccination, antibiotics, and stress reduction	Austin *et al.,* 2016
Flavobacteriosis (Columnaria or Bacterial Fin Rot)	*Flavobacterium columnare*	Freshwater fish (tilapia, carp, trout, catfish)	Fin rot, yellowish lesions on the skin and gills, and breathing difficulties.	Contaminated water, stress, and skin injuries.	Pond disinfection, antibiotics, and water quality improvement	Declercq *et al.,* 2013
Bacterial Hemorrhagic Septicemia (BHS)	*Aeromonas hydrophila*	Tilapia, Carp, Catfish, Trout	Skin haemorrhages, exophthalmos, anaemia, and ascites (abdominal swelling).	Contaminated water, injuries, stress.	Antibiotics, vaccination, and water quality monitoring.	Roberts, 2012

Pseudotuberculosis (Pasteurellosis of Fish)	*Photobacterium damselae subsp. piscicida*	Sea Bass, Sea Bream, Trout	Formation of white nodules in internal organs, weight loss, and high mortality.	Contaminated water, contact with infected fish.	Antibiotics, strict biosecurity, and vaccination	Magarinos *et al.*, 1996
Yersiniosis (Bacterial Kidney Disease of Salmonids)	*Yersinia ruckeri*	Rainbow trout, salmon	Kidney necrosis, cutaneous and internal haemorrhages, exophthalmos.	Contaminated water, contact with infected faeces	Vaccination, antibiotics, and stress reduction	Barnes, 2011

Table 11 Common fungal diseases of farmed fish

Disease	Pathogen	Affected Species	Symptoms	Transmission	Treatment and Prevention	Reference
Saprolegnia (Freshwater Fungus)	*Saprolegnia spp.* (mainly *Saprolegnia parasitica*)	Salmonids (trout, salmon), carp, tilapia, catfish	White, cottony patches on the skin, fins, and gills. Ulcers and tissue necrosis Lethargical behaviour,	stress, injuries, poor water quality, low temperatures (<15°C).	Improve water quality Treatment with salt, methylene blue, or hydrogen peroxide. Stress managemen	Van Den Berg *et al.*, 2013

			loss of appetite.		t and injury reduction.	
Branchiomycosis (Fungal Gill Disease)	*Branchiomyces sanguinis, Branchiomyces demigrans*	Carp, tilapia, catfish, sturgeon	Necrotic lesions on the gills. Rapid and laboured breathing. Sudden mortality without obvious external signs.	High temperatures, high organic load in the water, and stress.	Improve water oxygenation Clean and disinfect ponds. Use of antifungal treatments such as potassium permanganate.	Czeczuga and Muszynska, 1999
Ichthyophonosis (Fungal Granuloma Disease)	*Ichthyophonus hoferi*	Trout, salmon, herring, cod	Whitish nodules on the liver, kidneys, spleen, and muscles. Weight loss, lethargy, abnormal movement. Deformity of the spine in advanced cases.	Introduction of infected fish, chronic stress	Lack of effective treatment Avoid the introduction of carrier fish. Strict monitoring of farms.	Kocan *et al.*, 2004
Fish Fusarium Disease	*Fusarium spp.*	Tilapia, Trout, Catfish	Deep ulcerative lesions on the skin.	Poor hygiene, injuries, and environmental stress	Antifungal treatments. Rigorous pond cleaning.	Alves *et al.*, 2002

			Discolouration of fins and scales. Systemic infections leading to mortality.		Monitoring of water parameters.	

Table 12 Common Parasitic Diseases of Fishes

Disease	**Pathogen**	**Affected Species**	**Symptoms**	**Transmission**	**Treatment and Prevention**	**Reference**
Ichthyophthiriosis (White Spot Disease)	*Ichthyophthirius multifiliis* (freshwater fish) / *Cryptocaryon irritans* (marine fish)	Tilapia, trout, carp, catfish, sea bream, sea bass	Small white spots on the skin, fins, and gills. Rubbing against the pool walls. Accelerated breathing and loss of appetite.	Contaminated water, infected fish	Raising water temperature (>26°C) to disrupt the parasite's (*Ichthyophthirius*) life cycle. Treatments with salt, methylene blue, or formalin. Controlling fish density and improving water quality.	Buchmann and Bresciani, 2006
Trichodinosis	*Trichodina spp.*	Carp, Tilapia, Catfish, Trout	Skin and gill irritation. Excessive mucus secretion.	Contaminated water, infected fish	Salt or formalin bath. Improve water filtration and oxygenation.	Lom and Dykovà, 1992

			Abnormal swimming and lethargy.		Control stocking densities.	
Costiosis (Veil Disease)	*Ichthyobodo necator*	Trout, Tilapia, Catfish, Carp	Cloudy skin covered with a mucous veil. Loss of appetite and erratic swimming. Difficulty breathing and inflamed gills.	Contaminated water, infected fish.	Salt or formalin bath Improve husbandry conditions. Quarantine of new fish.	Woo, 2006
Gyrodactylosis and Dactylogyrosis (Monogenous Gill and Skin Worms)	*Gyrodactylus spp.* (skin) / *Dactylogyrus spp.* (gills)	Trout, Carp, Tilapia, Catfish	Rubbing against the walls. Rapid breathing and pale gills. Weight loss and high mortality in severe cases.	Direct contact between fish and contaminated water.	Antiparasitic bath (formalin, salt, praziquantel) Cleaning and disinfection of ponds Reducing fish stress	Bakke *et al.*, 2007

Lernaea (Anchorworm Disease)	*Lernaea spp*. (parasitic copepods)	Carp, Tilapia, Catfish	Parasitics visible as filaments attached to the body Ulcers and secondary infections. Weight loss and high mortality in severe infestations.	Contaminated water, infected fish	Manual removal of parasites. Treatments with diflubenzuron or organophosphates Pond disinfection and control of parasitic crustaceans.	Lester and Hayward, 2006
Myxosporidiosis	*Myxobolus spp., Henneguya spp.*	Salmonids, Carp, Tilapia	White cysts in the muscles, gills, or internal organs. Body deformity and abnormal swimming. Weight loss and reduced growth.	Infectious spores in the water, intermediate hosts (tubifex worms).	No effective treatment. Improvement of hygiene and husbandry conditions. Reduction of exposure to intermediate hosts.	Lom and Dykovà, 1992

Table 13 Common Protozoan Diseases

Disease	Pathogen	Affected Species	Symptoms	Transmission	Treatment and Prevention	Reference
Ichthyophthiriosis (White Spot Disease)	*Ichthyophthirius multifiliis* (freshwater) / *Cryptocaryon irritans* (marine water)	Trout, carp, tilapia, catfish, sea bream, sea bass.	Small white spots on the skin, fins, and gills. Rubbing against pond walls. Rapid breathing and loss of appetite.	Contaminated water, direct contact with infected fish.	Increase water temperature (>26°C) to accelerate the parasite's life cycle. Treatments with salt, methylene blue, or formalin. Monitoring and reduction of fish density.	Buchmann and Bresciani, 2006
Costiosis (Veil Disease)	*Ichthyobodo necator*	Trout, Tilapia, Carp, Catfish	Cloudy skin with excess mucus. Lethargy and loss of appetite. Inflamed gills and difficulty breathing.	Contaminated water, contact with infected fish.	Salt or formalin bath. Improve water and filtration conditions. Quarantine new fish.	Woo, 2006
Trichodinosis	*Trichodina spp.*	Carp, tilapia, catfish, trout	Rubbing against pond walls.	Contaminated water, contact with	Salt, formalin, or potassium permanganate bath.	Lom and Dykovà, 1992

			Excessive mucus secretion. Abnormal swimming and breathing difficulties.	infected fish.	Improve water oxygenation and filtration. Reduce stocking densities.	
Chilodonellosis	*Chilodonella spp.*	Carp, tilapia, trout, catfish	Bluish or greyish skin covered with excess mucus. Rapid breathing and difficulty breathing. Lethargy and disoriented swimming.	Contaminated water, contact with infected fish.	Salt or formalin treatments Improve hygiene conditions and water quality. Monitor fish when introducing new individuals.	Basson and Van As, 2006
Hexamitosis (Infection by Hexamita and Spironucleus)	*Hexamita spp., Spironucleus spp.*	Tilapia, trout, salmon, ornamental fish (discus)	Weight loss and loss of appetite. Whitish, gelatinous dropping. Intestinal lesions and high	Contaminated water, contact with infected feces.	Metrodinazole (antiparasitic) in feed. Improve water and feed conditions. Quarantine suspect fish.	Poynton and Morrison, 1990

			mortality in cases of severe infection.			
Myxosporidiosis (Infections by Myxobolus, Henneguya, etc.)	*Myxobolus spp., Henneguya spp.*	Carp, Salmon, Tilapia, Trout	Formation of white cysts on gills, muscles, or internal organs. Body deformities and slowed growth. Increased mortality in young fish.	Spores are released into the water and intermediate hosts, such as tubifex worms.	No effective treatment. Improved filtration and reduced exposure to intermediate hosts. Monitoring and controlling worm populations in ponds.	Lom and Dykovà, 2006

Table 14 Viral diseases

Disease	**Pathogen**	**Affected Species**	**Symptoms**	**Transmission**	**Treatment and Prevention**	**Reference**
Infectious Hematopoietic Necrosis (IHN)	Infectious Hematopoietic Necrosis Virus (IHNV), family Rhabdoviridae	Salmonids (rainbow trout, salmon).	Haemorrhages in the gills, skin, and fins. Disturbed swimming and loss of coordination.	Contaminated water, contact with infected fish, unsanitised fish farming equipment.	No specific treatment. Vaccination possible for certain species. Strengthened	Wolf, 1988

			Bulging eyes (exophthalmos) and swollen abdomen.		biosecurity and strict water quality control.	
Viral Haemorrhagic Septicemia (VHS)	Viral Haemorrhagic Septicemia Virus (VHSV), family Rhabdoviridae	Trout, salmon, perch, pikeperch, cod	Skin and internal haemorrhages. Abnormal swimming and loss of balance. Swollen abdomen and exophthalmos.	Contaminated water, contact with infected fish, and mechanical vectors (birds, equipment).	No effective treatment Strict health surveillance and elimination of infected fish. Vaccination in development.	WOAH, 2019
Infectious Pancreatic Necrosis (IPN)	Infectious Pancreatic Necrosis Virus (IPNV), family Birnaviridae	Trout, salmon, catfish, tilapia	Loss of appetite and slowed growth. Swollen abdomen and spiral swimming. High mortality in fry	Contaminated eggs, contact with infected fish, and contaminated water.	Lack of effective treatment Strict control of broodstock and eggs. Biosecurity and health surveillance	Roberts, 2012
Lymphocystosis	Lymphocystis Virus (LCV), family Iridoviridae	Marine and freshwater fish (sea bass, sea	Whitish or grey nodules on the skin and fins. Fin deformation.	Direct contact with infected fish, contaminated water.	No specific treatment. Isolation of infected fish and improveme	Chinchar, 2002

		bream, carp, tilapia)	Little impact on mortality, but it reduces the commercial value of the fish.		nt of water quality. Reduction of fish stress to prevent the onset of the disease.	
Viral Nervous Necrosis (VNN) or Viral Encephalopathy and Retinopathy (VRE)	Nodavirus, family Nodaviridae	Sea bass, sea bream, turbot, cod, halibut, tilapia	Loss of coordination and erratic movements. Skin discolouration. Significant mortality in larvae and young fish.	Contaminated water, vertical transmission (from parents to eggs)	No specific treatment Disinfection of eggs and strict control of broodstock. Elimination of infected individuals to limit the spread.	Munday *et al.*, 2002
Koi Herpesvirus (KHV)	*Cyprinid herpesvirus 3* (CyHV-3)	Common Carp, Koi Carp	Skin lesions and necrotic gills. Rapid breathing and restless behaviour. Very high mortality rates (80-100%) are expected in the event of an outbreak.	Direct contact with infected fish, contaminated water	No effective therapy. Strict health surveillance and quarantine of new fish. Development of experimental vaccines.	Haenen *et al.*, 2004

Disease Reporting

Reporting fish diseases is an essential step in preventing the spread of infections and protecting the health of aquaculture stocks, biodiversity, and public health. Several international and national organisations regulate disease reporting to ensure effective surveillance.

Disease reporting is important for:

- Prevention and control of epidemics.
- Protection of trade and exports.
- Health surveillance and biosecurity in aquaculture.
- Preservation of aquatic ecosystems and human health (WOAH, 2021).

Antimicrobial Resistance

Antimicrobial resistance (AMR) is a growing threat in aquaculture and aquatic ecosystems. The excessive or inappropriate use of antibiotics to treat bacterial diseases in fish promotes the development of resistant bacteria, compromising treatment effectiveness and posing risks to human and environmental health (WHO, 2021).

Although AMR develops naturally over time, antimicrobial misuse and overuse in humans and animals remain a major predisposing factor (Cabello, 2006; Chowdury *et al.,* 2022). This inappropriate use is linked to a lack of AMR and antimicrobial stewardship awareness and a lack of diagnostic capacity (mostly in low- and middle-income countries (LMICs). This affects proper identification of causative pathogens in diseased animals and antimicrobial prescriptions (Henriksson *et al.,* 2018; Adekanye *et al.,* 2020).

Another contributing factor is the use of antibiotics as prophylactics in disease prevention, especially in intensive factory farm settings of aquaculture production (Cabello, 2006). Furthermore, intensive aquaculture, poor animal welfare practices, and inadequate biosecurity can increase the risk of infection in fish and consequently lead to increased antibiotic use (Cabello, 2006).

Antibiotics are typically administered to fish through feeds, in baths, or via injections (Chowdury *et al.,* 2022). These methods can lead to the accumulation of antibiotic residues in the fish and their aquatic ecosystems. If the proper withdrawal periods are not observed after administering antibiotics, consumers of such fish will ingest antibiotic residues at suboptimal doses, which can facilitate the development of AMR and other health risks (Heuer *et al.,* 2009; Sapkota *et al.,* 2008). Furthermore, these residues and resistant bacteria can be transferred between the aquatic and terrestrial animals through the environment and waterways (Goldburg and Naylor, 2005; Naylor and Burke, 2005; Chowdury *et al.,* 2022).

How does AMR spread from animals to humans?

Antimicrobial resistance (AMR) primarily spreads from animals to humans through food, the environment, and direct contact with contaminated animals or their products. This phenomenon is particularly worrying in aquaculture, where excessive use of antibiotics promotes the emergence of resistant bacteria that can infect humans (Akinbowale *et al.,* 2006).

Impact of AMR

Antimicrobials are essential in intensive animal agriculture and aquaculture. Antibiotics, including oxytetracycline, amoxicillin, and sulphadiazine trimethoprim, are used extensively in aquaculture to treat or prevent fish diseases, thus maximising productivity (Chowdury *et al.,* 2022). However, misuse and overuse lead to AMR, which causes treatment failure and affects aquaculture fish production and welfare (Schar *et al.,* 2020).

Furthermore, antimicrobial misuse in aquaculture results in wide contamination of the environment with antimicrobial residues via water distribution systems (Schar *et al.,* 2020). These residues can affect the environment's microbiome and, consequently, its regulatory and supporting activities in ecosystems (Sarmah *et* *al.,* 2006; Larsson *et al.,* 2018). Also, aquaculture systems with high antimicrobial use may serve as reservoirs for antimicrobial resistance genes, hence facilitating AMR development in animals and humans (Schar *et al.,* 2020). We should also consider that authorised antibiotics for aquaculture species are scarce globally; hence, their efficacies should be maintained.

Combatting AMR

Combating antimicrobial resistance (AMR) is a global priority to protect human, animal, and environmental health. In aquaculture, where the excessive use of antibiotics promotes the emergence of resistant bacteria, it is essential to adopt sustainable strategies to limit this phenomenon.

The FAO action plan on AMR (2016–2020) recommends the prudent use of antimicrobials and effective biosecurity practices (FAO). The main recommendations include:

1. Prudent and responsible use of antimicrobials to preserve their efficacy.
2. Provision of clean, safe, and disease-free aquatic systems to prevent infectious disease incidence and reduce antimicrobial use.
3. Proper routine monitoring of resistance during disease outbreaks.
4. Proper animal welfare standards should be adopted and maintained as they ensure better immune systems in animals, thus preventing infections, minimising outbreaks, and reducing antimicrobial use.
5. Routine removal of antibiotic residues in water via appropriate adsorption techniques, filtration, biological methods, sedimentation, and flocculation (Homem and Santos, 2011).
6. Vaccination of aquatic food animals for infectious disease prevention. For example, oral fish vaccines are effective against many aquatic diseases (Newaj-Fyzul and Austin, 2015).
7. Probiotics should also be considered in infection prevention and control. For example, probiotics are potential alternatives in controlling pathogens

such as *Vibrio harveyi*, a major health threat in aquaculture (Chabrillon *et al.,* 2005).

8. Immunostimulants can also be considered for use. An example is β-1,3 glucans, which are reportedly effective alternatives against various aquatic diseases like *vibriosis*, enteric redmouth, *aeromonadiasis*, *pasteurellosis*, and Hitra disease (Ngamkala *et al.,* 2010).
9. Broad-host range phages can also be considered to treat bacterial infections. For example, due to the unavailability of appropriate vaccines, phages were used in salmonids to prevent rainbow trout fry syndrome (RTFS) caused by *Flavobacterium psychrophilum* (Castillo *et al.,* 2012).
10. Traditional medicinal plants can also be explored as antimicrobial alternatives. Examples include seaweeds, extracts of mango, peppermint, turmeric, jasmine, and neem, which are promising alternatives for treating bacterial infections caused by aeromonads and vibrios in aquatic animals (Newaj Fyzul and Austin, 2015).

The fight against antimicrobial resistance (AMR) in aquaculture relies on the responsible use of antibiotics, the development of sustainable alternatives, strict regulation, and increased awareness among industry stakeholders.

An integrated and coordinated approach involving producers, scientists, health authorities, and consumers is essential to reduce the spread of AMR and ensure sustainable aquaculture that respects public health.

Q&A Session

- What is antimicrobial resistance (AMR) and why is it a threat to public health and aquaculture?
- What are the main factors promoting the emergence of AMR in aquaculture?
- What types of microorganisms can develop resistance to antimicrobials?
- What are the main modes of transmission of AMR from fish to humans?

REFERENCES

Abecasis, D., and Erzini, K. (2008). Site fidelity and movements of gilthead sea Coastal bream (*Sparus aurata*) in a coastal lagoon (Ria Formosa, Portugal). *Estuarine, and Shelf Science*, 79(4), 758–763. https://doi.org/10.1016/j.ecss.2008.06.019.

Adekanye, U. O., Ekiri, A. B., Galipó, E., Muhammad, A. B., Mateus, A., La Ragione, R. M., ... and Cook, A. J. (2020). Knowledge, attitudes and practices of veterinarians towards antimicrobial resistance and stewardship in Nigeria. *Antibiotics*, 9(8), 453. https://doi.org/10.3390/antibiotics9080453.

Akinbowale, O. L., Peng, H., and Barton, M. D. (2006). Antimicrobial resistance in bacteria isolated from aquaculture sources in Australia. *Journal of Applied Microbiology*, 100(5), 1103-1113.

Animal Welfare Institute. (2018). The critical relationship between farm animal health and welfare. Available from: https://awionline.org/sites/default/files/uploads/documents/FA-AWI-Animal-Health_Welfare-Report-04022018.pdf. Animals kept under Intensive Livestock Husbandry Systems. Her Majesty's Stationery Office. London

Arechavala-Lopez, P., Caballero-Froilán, J. C., Jiménez-García, M., Capó, X., Tejada, S., Saraiva, J. L., Sureda, A., and Moranta, D. (2020). Enriched environments enhance cognition, exploratory behaviour and brain physiological functions of *Sparus aurata*. *Scientific Reports*, 10(1), 11252. https://doi.org/10.1038/s41598-020-68306-6.

Arechavala-Lopez, P., Cabrera-Álvarez, M. J., Maia, C. M., and Saraiva, J. L. (2021). Environmental enrichment in fish aquaculture: A review of fundamental and practical aspects. *Reviews in Aquaculture*, 14(2). https://doi.org/10.1111/raq.12620.

Arechavala-Lopez, P., Diaz-Gil, C., Saraiva, J.L., Moranta, D., Castanheira, M.F., Nuñez-Velazquez, S., Ledesma-Corvi, S., Mora-Ruiz, M.R., Grau, A. (2019). Effects of structural environmental enrichment on juvenile seabream (*Sparus aurata*) under feed deficit conditions. *Applied Animal Behaviour Science*, submitted.

Ashley, P. J. (2007). Fish welfare: Current issues in aquaculture. Applied Animal Behaviour Science, 104(3-4), 199–235. DOI: 10.1016/j.applanim.2006.09.00.

Austin, B., & Austin, D. A. (2016). *Bacterial Fish Pathogens: Disease of Farmed and Wild Fish*. Springer.

Avnimelech, Y. (2012). *Biofloc Technology: A Practical Guide Book*. The World Aquaculture Society.

Bakke, T. A., Harris, P. D., & Cable, J. (2007). The biology of *Gyrodactylid* monogeneans: the "Russian-doll killers". *Advances in Parasitology*, 64, 161-376.

Barnes, A. C. (2011). Enteric redmouth disease (ERM) (Yersinia ruckeri). In *Fish Diseases and Disorders* (Vol. 3, pp. 484-511).

Barreto, R. E., Carvalho, G. G. A., and Volpato, G. L. (2011). The aggressive behaviour of Nile tilapia introduced into novel environments with variation in enrichment. *Zoology*, 114(1), 53–57. https://doi.org/10.1016/j.zool.2010.09.001.

Basson, L., and Van As, J. G. (2006). Chilodonellosis in freshwater fishes. *Journal of Fish Diseases*, 29(6), 425-431.

Batzina, A., and Karakatsouli, N. (2012). The presence of substrate as a means of environmental enrichment in intensively reared gilthead seabream *Sparus aurata*: growth and behavioural effects. *Aquaculture*, 370–371, 54–60. https://doi.org/10.1016/J.AQUACULTURE.2012.10.005

Batzina, A., and Karakatsouli, N. (2014). Is it the blue gravel substrate or only its blue colour that improves growth and reduces aggressive behaviour of gilthead seabream *Sparus aurata*? *Aquacultural Engineering*, 62:49-53.

Batzina, A., Dalla, C., Papadopoulou-Daifoti, Z., and Karakatsouli, N. (2014a). Effects of environmental enrichment on growth, aggressive behaviour and brain monoamines of gilthead seabream *Sparus aurata* reared under different social conditions. *Comparative Biochemistry and Physiology Part A: Molecular and Integrative Physiology*, 169:25-32.

Batzina, A., Dalla, C., Tsopelakos, A., Papadopoulou-Daifoti, Z., and Karakatsouli, N. (2014b). Environmental enrichment induces changes in brain monoamine levels in gilthead seabream *Sparus aurata*. *Physiology and Behaviour*, 130:85-90.

Batzina, A., Kalogiannis, D., Dalla, C., Papadopoulou-Daifoti, Z., Chadio, S., and Karakatsouli, N. (2014c). Blue substrate modifies the time course of stress response in gilthead seabream *Sparus aurata*. *Aquaculture*, 420:247-253.

Batzina, A., Sotirakoglou, K., and Karakatsouli, N. (2014d). The preference of 0+ and 2+ gilthead seabream Sparus aurata for coloured substrates or no-substrate. Applied *Animal Behaviour Science*, 151:110-116.

Bera, K.K., Karmaka, S., Jana, P., Das, S. K. Purkait, S., Pal, S., and Haque, R. (2018). Biosecurity in aquaculture: An overview. *Aqua International*, 42.

Bosakowski, T., and Wagner, E. J. (1995). Experimental use of cobble substrates in concrete raceways for improving fin condition of cutthroat (*Oncorhynchus clarki*) and rainbow trout (O. mykiss). *Aquaculture*, 130(2–3), 159–165.

Boyd, C.E. (1998). Water quality for pond aquaculture. *Research and Development* Series No. 43. Auburn University.

Boyd, C.E. *et al.* (2016). *Water quality in aquaculture: managing high-density fish production*.

Boyd, C.E., and Tucker, C.S. (1998). *Pond aquaculture water quality management*. Springer.

Boyd, C.E., Tucker, C.S., and Somridhivej, B. (2020). Aquaculture and the environment: Towards sustainability. *Annual Review of Environment and Resources*, 45(1), 229–252. DOI: 10.1146/annurev-environ-012420-05000.

Braithwaite, V. A., Huntingford, F. A., and van den Bos, R. (2006). Variation in Emotion and Cognition Among Fishes. *Journal of Fish Biology*, 68(2), 489-493.

Brambell, R. (1965). Report of the Technical Committee to Enquire into the Welfare of Animals kept under Intensive Livestock Husbandry Systems. Her Majesty's Stationery Office. London.

Brett, J. R., and Groves, T. D. D. (1979). Physiological energetics. *Fish Physiology*, 8, 279-352.

Brockmark, S., Neregård, L., Bohlin, T., Björnsson, B.T., and Johnsson, J.I. (2007). Effects of rearing density and structural complexity on the pre- and post-release performance of Atlantic salmon. *Transactions of the American Fisheries Society*, 136(5), 1453–1462. https://doi.org/10.1577/t06-245.1.

Broom, D.M. (2011). A history of animal welfare science. *ActaBiotheoretica*, 2/59, 121-137. DOI: 10.1007/s10441-011-9123-3.

Brown, C., Laland, K., and Krause, J. (Eds.). (2011). *Fish Cognition and Behaviour.* Wiley-Blackwell.

Buchmann, K., and Bresciani, J. (2006). *Ichthyophthirius multifiliis* (Fish "White Spot" Disease). *Journal of Fish Diseases*, 29(8), 561-574

Bunting, S. W., and Pretty, J. (2007). Aquaculture development and global carbon budgets: Emissions, sequestration, and management options. *Science of the Total Environment*, 374(2-3), 313-332.

Cabello, F. C. (2006). Heavy use of prophylactic antibiotics in aquaculture: a growing problem for human and animal health and for the environment. *Environmental microbiology*, 8(7), 1137-1144. https://doi.org/10.1111/j.1462-2920.2006.01054.x.

Cascarano, M.C., Stavrakidis-Zachou, O., Mladineo, I., Thompson, K.D., Papandroulakis, N., and Katharios, P. (2021). Mediterranean aquaculture in a changing climate: temperature effects on pathogens and diseases of three farmed fish species. *Pathogens*, 10(9), 1205. https://doi.org/10.3390/pathogens10091205.

Castillo, D., Higuera, G., Villa, M., Middelboe, M., Dalsgaard, I., Madsen, L., and Espejo, R. T. (2012). Diversity of *Flavobacterium psychrophilum* and the potential use of its phages for protection against bacterial cold water disease in salmonids. *Journal of Fish Diseases*, 35(3), 193-201. https://doi.org/10.1111/j.1365-2761.2011.01336.x.

Chabrillón, M., Rico, R.M., Arijo, S., Díaz-Rosales, P., Balebona, M.C., and Moriñigo, M.A. (2005). Interactions of microorganisms isolated from gilthead sea bream, *Sparus aurata* L., on *Vibrio harveyi*, a pathogen of farmed Senegalese sole, *Solea senegalensis* (Kaup). *Journal of Fish Diseases, 28(9), 531*- 537. https://doi.org/10.1111/j.1365-2761.2005.00657.x.

Chinchar, V. G. (2002). Ranaviruses (family Iridoviridae): emerging cold-blooded killers. *Archives of Virology*, 147(3), 447-470.

Chowdhury, S., Rheman, S., Debnath, N., Delamare-Deboutteville, J., Akhtar, Z., Ghosh, S., Parveen, S., Islam, K., Islam, M. A., Rashid, M. M., Khan, Z. H., Rahman, M., Chadag, V. M., and Chowdhury, F. (2022). Antibiotic usage practices in aquaculture in Bangladesh and their associated factors. One

Health (Amsterdam, Netherlands), 15, 100445. https://doi.org/10.1016/j.onehlt.2022.100445.
Chulayo A.Y., Muchenje V. (2015). A balanced perspective on animal welfare for improved meat and meat products. S Afr J AnimSci2015; 45:452-69.
Clottey J.A. (1985). Slaughter practices and techniques. In: Manual for the slaughter of small ruminants in developing countries. FAO Animal Production and Health Paper 49. Rome, Italy: FAO.
Conte, F.S. (2004). Stress and the Welfare of Cultured Fish. *Applied Animal Behaviour Science*, 86(3-4), 205–223.
Czeczuga, B., and Muszyńska, E. (1999). Occurrence of *Branchiomyces* species in fish ponds with environmental conditions. *Acta Mycologica*, 34(2), 209-217.
Declercq, A.M., Haesebrouck, F., Van den Broeck, W., Bossier, P., and Decostere, A. (2013). Columnaris disease in fish: a review with emphasis on bacterium-host interactions. *Veterinary Research*, 44(1), 27.
Degen AA. (2007). Sheep and goat milk in pastoral societies. *Small RuminRes2007*; 68:7-19.
Direction générale de la pêche et de l'Aquaculture (DGAP). (2023). Annuaire des statistiques des pêches en Tunisie. Direction générale de la pêche et de l'Aquaculture : 30 p.
Edwards, P. (2000). Aquaculture, poverty impacts, and livelihoods. Natural Resource Perspectives, 56.
EFSA. (2017). Welfare of Farmed Fish: Common Practices during Transport and at Slaughter. https://ec.europa.eu/food/sites/food/files/animals/docs/aw_platform_20180621_pre-06.pdf.
El-Sayed, A. F. M. (2006). *Tilapia Culture*. CABI Publishing.
Espinosa, M.A., García-Gallego, M., and Hidalgo, M.C. (2015). *Biology and culture of European Seabass* (*Dicentrarchus labrax*). CRC Press.
FAO (Organisation des Nations Unies pour l'Alimentation et l'Agriculture). (2020). Guide sur l'aquaculture durable et le bien-être des poissons.
FAO, rapports sur la pêche méditerranéenne et études tunisiennes.
Fishcount. (2019). Number of farmed fish slaughtered each year | fishcount.org.uk. Fishcount.org.uk. http://fishcount.org.uk/fish-count-estimates-2/numbers-of-farmed-fish-slaughtered-each-year.
Fraser, D. (2008). *Understanding animal welfare: The science in its cultural context*. Wiley-Blackwell.
Goldburg, R., and Naylor, R. (2005). Future seascapes, fishing, and fish farming. *Frontiers in Ecology and the Environment*, 3(1), 21-28. https://doi.org/10.1890/1540-9295(2005)003%5b0021:FSFAFF%5d2.0.CO;2.
Grandin T. (2018). Welfare problems in cattle, pigs, and sheep that persist even though scientific research clearly shows how to prevent them. *Animals* 2018;8:124. https://doi.org/10.3390/ani8070124.

Gray S, Sundal M, Wiebusch B, Little M.A., Leslie P.W., Pike I.L. (2003). Cattle raiding, cultural survival, and the adaptability of East African pastoralists. CurrAnthropol 44(5 suppl.);2003:S3-30. https://doi.org/10.1086/377669.

Haenen, O.L.M., Way, K., Bergmann, S.M., and Ariel, E. (2004). The emergence of koi herpesvirus (KHV) and its significance to European aquaculture. *Bulletin of the European Association of Fish Pathologists*, 24(6), 293-307.

Hambrey, J., and Evans, S. (2016). Aquaculture and community development. *World Aquaculture Society Reports.*

Harmon, T. S. (2009). Methods for reducing stressors and maintaining fish health during live fish transport. *North American Journal of Aquaculture*, 71(3), 262-277.

Henriksson, P. J., Rico, A., Troell, M., Klinger, D. H., Buschmann, A. H., Saksida, S., ... and Zhang, W. (2018). Unpacking factors influencing antimicrobial use in global aquaculture and their implication for management: a review from a systems perspective. *Sustainability Science,* 13, 1105-1120. https://doi.org/10.1007/s11625-017-0511-8.

Henry, M., Gasco, L., Piccolo, G., and Fountoulaki, E. (2015). Insect meal as an alternative source of protein in aquaculture diets: A review. *Aquaculture*, 437, 1-14.

Herrero M, Grace D, Njuki J, *et al.* (2013). The roles of livestock in developing countries. *Animal* 2013;7(Suppl 1):3-18. https://doi.org/10.1017/S1751731112001954.

Heuer, O.E., Kruse, H., Grave, K., Collignon, P., Karunasagar, I., and Angulo, F.J. (2009). Human health consequences of the use of antimicrobial agents in aquaculture. *Clinical Infectious Diseases,* 49(8), 1248 1253. https://doi.org/10.1007/s11625-017-0511-8.

Hoffman, I. (2010). Climate change and the characterisation, breeding and conservation of animal genetic resources. *Anim Genet 2010*;41:32-46. https://doi.org/10.1111/j.1365-2052.2010.02043.x.

Holmyard, N. (2017). Fish producers benefit from humane slaughter techniques - Responsible seafood advocate. *Global Seafood Alliance.* https://www.globalseafood.org/advocate/fish-producers-benefit humane-slaughter-techniques/.

Homem, V., & Santos, L. (2011). Degradation and removal methods of antibiotics from aqueous matrices–a review. *Journal of Environmental Management*, 92(10), 2304-2347. https://doi.org/10.1016/j.jenvman.2011.05.023.

https://aquaculturefrance.com/fr/accueil/467-oxyguard-pacific.

https://asc-aqua.org/blog/explained-what-is-ras-aquaculture/

https://doi.org/10.1016/j.livsci.2011.07.014.

https://gipp.tn/fr/fili%C3%A8res/daurade-et-loup-daquaculture

https://inspection.canada.ca/fr/exigences-documents-dorientation-relatives-c/produits-viande-animaux-alimentation-hu/methodes-dabattage-surveillance.

https://chaire-bea.vetagro-sup.fr/pourquoi-et-comment-enrichir-lenvironnement-des-animaux-delevage

https://macommande.lemonde.fr/account/create/ESSADIMENS1299PRIO/ESS365J14199PRLVT1FPRIO?marketing=241RESSCMP.

https://welfarm.fr/nos-champs-d-action/labattage-des-animaux-delevage/

https://www.agrociwf.fr/media/7441968/ciwf-bar-et-daurade-ameliorer-labattage-des-bars-et-daurade.pdf

https://www.aquaportail.com/dictionnaire/definition/6595/raceway.

https://www.ctaqua.tn/2021/04/07/la-peche-en-tunisie/

https://www.fao.org/fishery/docs/CDrom/aquaculture.

https://www.l214.com/animaux/abattage/abattage-sans-etourdissement

https://www.linde.ch/shop/fr/ch-ig/oxygenation-en-aquaculture?

https://www.publications.gov.on.ca/store/20170501121/Free_Download_Files/301648.pdf

https://www.wcl.org.uk/duplicate-of-moving-to-the-five-domains-model-for-assessing-animal-welfare.asp

Humane Slaughter Association. (2004). Humane harvesting of farmed fish. Guidance notes No. 5. Humane Slaughter Association, Wheathampstead, Hertfordshire. 1-23. https://www.hsa.org.uk/downloads/publications/harvestingfishdownload-updated-with-2016-logo.pdf.

Huntingford, F.A., Adams, C., and Kadri, S. (2006). Behavioural Responses to Stress in Fish: A Review. *Journal of Fish Biology*.

Huntingford, F.A., Adams, C., Braithwaite, V.A., *et al.* (2006). Current issues in fish welfare. *Journal of Fish Biology*, 68(2), 332–372. DOI: 10.1111/j.0022-1112.2006.001046.x.

Jobling, M., and Peruzzi, S. (2010). Seabreams and Porgies (Family: Sparidae). In N.R. Le Francois, M. Jobling, C. Carter, & P. Blie (Eds.), Finfish Aquaculture Diversification(pp.361-373).CABI Publishing. https://books.google.be/books?hl=en&lr=&id=rZL_d2lyUsgC&oi=fnd&pg=PA361&dq=Jobling,+Malcolm,+and+Stefano+Peruzzi.+2010.+Seabreams+and+Porgies+(Family:+Sparidae).+In+Finfish+Aquaculture+Diversification,+ed.+Nathalie+R.+Le+Francois,+Malcolm+Jobling,+Chris+C.

Kaushik, S. J., and Troell, M. (2010). Feeding efficiency and sustainability in aquaculture. *Reviews in Aquaculture,* 2(3), 117-124.

Kocan, R.M., Hershberger, P.K., and Winton, J.R. (2004). Ichthyophoniasis: an emerging disease of Chinook salmon in the Pacific Northwest. *Journal of Aquatic Animal Health*, 16(2), 58-72.

Larsson, D.J., Andremont, A., Bengtsson-Palme, J., Brandt, K.K., de Roda Husman, A.M., Fagerstedt, P., ... and Wernersson, A.S. (2018). Critical knowledge gaps and research needs related to the environmental dimensions of antibiotic resistance. *Environment International*, 117, 132-138. https://doi.org/10.1016/j.envint.2018.04.041.

Leone, E.H., and Estévez, I. (2008). Economic and Welfare Benefits of Environmental Enrichment for Broiler Breeders. *Poultry Science*, 87(1), 14–21. https://doi.org/10.3382/ps.2007-00154.

Lester, R. J. G., and Hayward, C. J. (2006). Phylum Arthropoda. In *Fish Diseases and Disorders, Volume 1: Protozoan and Metazoan Infections*.

Lom, J., and Dyková, I. (1992). Protozoan parasites of fishes. *Elsevier Science Publishers B.V*.

Lom, J., and Dyková, I. (2006). *Myxozoan genera: definition and notes on taxonomy, life-cycle terminology and pathogenic species*. *Folia Parasitologica*, 53(1), 1-36.

Magariños, B., Romalde, J.L., and Toranzo, A.E. (1996). Pathogenicity and antigenic characterisation of *Pasteurella piscicida* strains isolated from fish. *Applied and Environmental Microbiology*, 62(11), 4074-4078. Mathilde Escudero. (2018). Bien-être des poissons en aquaculture. Médecine vétérinaire et santé animale. ffdumas-04885683f.

Mellor, D.J, and Beausoleil, N.J. (2015). Extending the 'Five Domains' model for animal welfare assessment to incorporate positive welfare states. *Animal Welfare*, 24(3), 241-253.

Merrifield, D.L., Dimitroglou, A., Foey, A., Davies, S.J., Baker, R.T.M., Bøgwald, J., ... and Ringø, E. (2010). The current status and future focus of probiotic and prebiotic applications for salmonids. *Aquaculture,* 302(1-2), 1-18.

Muchenje V, Mukumbo F.E., Njisane Y.Z. (2018). Meat in a sustainable food system. *S Afr J AnimSci2018*; 48:818-28. http://dx.doi.org/10.4314/sajas.v48i5.3.

Mukaila, R., Ukwuaba, I.C., & Umaru, I.I. (2023). Economic Impact of Disease on Small-Scale Catfish Farms in Nigeria. *Aquaculture*, 739773. https://doi.org/10.1016/j.aquaculture.2023.739773.

Munday, B.L., Kwang, J., and Moody, N. (2002). *Betanodavirus infections of marine fish*. *Annual Review of Fish Diseases*, 12, 41-64.

Näslund, J., and Johnson, J.I. (2014). Environmental enrichment for fish in captive environments: effects of physical structures and substrates. *Fish and Fisheries,* 17(1), 1–30. https://doi.org/10.1111/faf.12088.

Naylor, R., and Burke, M. (2005). Aquaculture and ocean resources: Raising Tigers of the sea. *Annual Review of Environment and Resources*, 30(1), 185–218. https://doi.org/10.1146/annurev.energy.30.081804.121034.

Ndou S.P., Muchenje V, Chimonyo M. (2011). Assessment and implications of animal welfare in beef production systems in developing countries. *Afr J Biotechnol 2011*;10:1049-64.

Neto, J.F. and Percilia, C.G. (2020). Environmental enrichment techniques and tryptophan supplementation used to improve the quality of life and animal welfare of Nile tilapia. *Aquaculture Reports*, 17: 100354.

Newaj-Fyzul, A., and Austin, B. (2015). Probiotics, immunostimulants, plant products and oral vaccines, and their role as feed supplements in the control of bacterial fish diseases. *Journal of Fish Diseases,* 38(11), 937-955. https://doi.org/10.1111/jfd.12313.

Ngamkala, S., Futami, K., Endo, M., Maita, M., and Katagiri, T. (2010). Immunological effects of glucan and *Lactobacillus rhamnosus* GG, a probiotic bacterium, on Nile tilapia *Oreochromis niloticus* intestine with oral Aeromonas challenges. *Fisheries Science*, 76, 833-840. https://doi.org/10.1007/s12562-010-0280-0.

Nicks, B., and Vandenheede, M. (2014). Animal health and welfare: equivalent or complementary?. *Revue scientifique et technique (International Office of Epizootics*), 33(1), 97–96. https://doi.org/10.20506/rst.33.1.2261. nie.com

Njisane Y.Z., Muchenje V. (2017). Farm to abattoir conditions, animal factors and their subsequent effects on cattle behavioural responses and beef quality — a review. *Asian-Australas J AnimSci2017*;30:755-64. https://doi.org/10.5713/ajas.16.0037.

Njisane Y.Z., Mukumbo F.E., and Muchenje V. (2020). An outlook on livestock welfare conditions in African communities — A review. *Asian-Australas J AnimSci* Vol. 33, No. 6:867-878 June 2020https://doi.org/10.5713/ajas.19.0282pISSN 1011-2367 eISSN 1976-5517.

NRC (2011). *Nutrient requirements of fish and shrimp*. National Research Council.

Nyika A. (2009). Animal research ethics in Africa: An overview. *Acta Trop 2009*;112 (Suppl 1):S48-52. https://doi.org/10.1016/j. actatropica.2009.07.021.

Papoutsoglou, S.E., Karakatsouli, N., Batzina, A., Papoutsoglou, E.S., and Tsopelakos, A. (2008). Effect of music stimulus on gilthead seabream *Sparus aurata* physiology under different light intensity in a re-circulating water system. *Journal of Fish Biology*, 73(4), 980–1004. https://doi.org/10.1111/j.1095-8649.2008.02001.x.

Papoutsoglou, Sofronios E., Karakatsouli, N., Skouradakis, C., Papoutsoglou, E.S., Batzina, A., Leondaritis, G., and Sakellaridis, N. (2013). Effect of musical stimuli and white noise on rainbow trout (*Oncorhynchus mykiss*) growth and physiology in recirculating water conditions. *Aquacultural Engineering, 55*, 16–22. https://doi.org/10.1016/j.aquaeng.2013.01.003.

Poli, B. M., Parisi, G., Scappini, F., and Zampacavallo, G. (2005). Fish welfare and quality as affected by pre-slaughter and slaughter management. *Aquaculture International*, 13(1-2), 29–49. https://doi.org/10.1007/s10499-004-9035-1

Poynton, S.L., and Morrison, C.M. (1990). Morphology of *Spironucleus* species (Diplomonadida, Hexamitidae) from fish. *Canadian Journal of Zoology*, 68(4), 667-685.

Rakocy, J.E., Masser, M.P., and Losordo, T.M. (2006). Recirculating aquaculture tank production systems: Aquaponics — Integrating fish and plant culture. Southern Regional Aquaculture Centre Publication, 454.

Ren, Y., Xiong, M., Yu, J., Li, W., Li, B., Liu, J., and Zhang, T. (2019). Effects of artificial submerged vegetation on consumption and growth of mandarin fish *Siniperca chuatsi* (Basilewsky) foraging on live prey. *Journal of Freshwater Ecology*, 34:433–444.

Roberts, L.J., Taylor, J., and de Leaniz, C.G. (2011). Environmental enrichment reduces maladaptive risk-taking behaviour in salmon reared for conservation. *Biological Conservation*, 144: 1972-1979.

Roberts, R.J. (2012). *Fish Pathology*. Wiley-Blackwell.

Ross, L.G., and Ross, B. (2008). *Anaesthetic and Sedative Techniques for Aquatic Animals*. Blackwell Publishing.

RSPCA (Royal Society for the Prevention of Cruelty to Animals). Guide sur les normes pour le bien-être des poissons. Disponible sur : https://www.rspca.org.uk/.

Sabri, B.D.M., Elnwishy, N., and Nwonwu, F. (2012). Effect of Environmental Colour on the Behavioural and Physiological Responses of Nile Tilapia, *Oreochromis niloticus*. *Global Journal of Science Frontier Research*, 12(4), 11–20.

Sapkota, A., Sapkota, A. R., Kucharski, M., Burke, J., McKenzie, S., Walker, P., and Lawrence, R. (2008). Aquaculture Practices and Potential Human Health Risks: Current Knowledge and Future Priorities. *Environment International*, 34(8), 1215-1226. https://doi.org/10.1016/j.envint.2008.04.009.

Sarmah, A.K., Meyer, M.T., and Boxall, A.B. (2006). A global perspective on the use, sales, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment. *Chemosphere*, 65(5), 725-759. https://doi.org/10.1016/j.chemosphere.2006.03.026

Schar, D., Klein, E.Y., Laxminarayan, R., Gilbert, M., and Van Boeckel, T.P. (2020). Global Trends in Antimicrobial Use in Aquaculture *Scientific Reports*, 10(1), 21878. https://doi.org/10.1038/s41598-020 78849-3.

Scholtz M.M., McManus C, Okeyo A.M., Theunissen A. (2011). Opportunities for beef production in developing countries of the southern hemisphere. *LivestSci2011*;142:195-202.

Soares, M.C., Oliveira, R.F., Ros, A.F.H., Grutter, A.S., and Bshary, R. (2011). Tactile stimulation lowers stress in fish. *Nature Communications,* 2(1), 1–5. https://doi.org/10.1038/ncomms1547.

Subasinghe, R., *et al.* (2019). *Health management in aquaculture: Practices and challenges*.

Subasinghe, R., Soto, D., and Jia, J. (2009). Global Aquaculture and Its Role in Sustainable Development. *Reviews in Aquaculture*, 1(1), 2-9. https://doi.org/10.1111/j.1753-5131.2008.01002.x.

Tacon, A.G., and Metian, M. (2008). Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: Trends and future prospects. *Aquaculture*, 285(1-4), 146-158.

Tacon, A.G.J., and Metian, M. (2015). *Feed Management and Nutrition in Aquaculture*. Springer.

Timmons, M.B., and Ebeling, J.M. (2010). *Recirculating aquaculture systems*. Cayuga Aqua Ventures.

Toranzo, A.E., Magariños, B., and Romalde, J.L. (2005). A review of the main bacterial fish diseases in mariculture systems. *Aquaculture*, 246(1-4), 37-61.

Torrezani, C.S., Pinho-Neto, C.F., Miyai, C.A., Fábio, e Barreto, R.E. (2013). Structural enrichment reduces aggression. *Tilapia rendalli*. 46(3), 183–190. https://doi.org/10.1080/10236244.2013.805053

WOAH (2019). *Septicémie hémorragique virale*, Manuel de l'OIE.

WOAH (2021). Animal Welfare and International Standards. Organisation mondiale de la santé animale.

Wolf, K. (1988). *Fish viruses and fish viral diseases*. Cornell University Press.

Woo, P.T.K. (2006). *Fish diseases and disorders, Volume 1: Protozoan and Metazoan Infections*. CABI Publishing.

Woodward, M., Winder, L., and Watt, P. (2019). Enrichment increases aggression in zebrafish. *Fishes*, 4(1), 22. https://doi.org/10.3390/fishes4010022

World Health Organisation (WHO). (2021). Antimicrobial resistance [Fact sheet]. Available from: https://www.who.int/news-room/fact-sheets/detail/antimicrobial-resistance.

www.oie.int

www.tunisie-aquapo

Yanong, R. P., and Erlacher-Reid, C. (2012). Biosecurity in aquaculture, part 1: An overview. *SRAC publication*, 4707, 522. http://fisheries.tamu.edu/files/2013/09/SRAC-Publication-No.-4707-Biosecurity-in Aquaculture-Part-1-An-Overview.pdf.

Zhang, Z., Bai, Q., Xu, X., Guo, H., and Zhang, X. (2020). Effects of environmental enrichment on the welfare of juvenile black rockfish Sebastes schlegelii: Growth, behaviour and physiology. *Aquaculture*, 518, 734782.https://doi.org/10.1016/j.aquaculture.2019.734782.

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