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# *Aquatropolis – Contributions for the definition of an intelligent management system in aquaculture.*

Simone Isabel Ferreira Teixeira

[2017]

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# *Aquatropolis – Contributions for the definition of an intelligent management system in aquaculture.*

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Dissertation for the Master's Degree in Aquaculture

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Title: Aquatropolis – Contributions for the definition of an intelligent management system in aquaculture.

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# Resumo

A pesca e a aquacultura continuam a ser fontes importantes de alimento, nutrição, rendimento e meios de subsistência para centenas de milhões de pessoas em todo o mundo. O abastecimento mundial de peixe *per capita* atingiu um novo recorde de 20 kg em 2014, graças ao vigoroso crescimento da aquacultura, que agora fornece metade de todo o peixe para consumo humano e a uma ligeira melhora no estado de determinadas unidades populacionais de peixes devido à melhoria da gestão da pesca. Atualmente, uma quarta parte do pescado consumido na UE é produzido em aquacultura, em 2011, 1,24 milhões de toneladas de pescado de aquacultura foram produzidos na UE, com um valor de € 3,51 bilhões. Os modelos de produção têm como objetivo o desenvolvimento de estratégias que ajudem a aquacultura a tornarem-se mais eficientes e menos nocivas para o meio ambiente, as espécies robalo e dourada são espécies representativas da aquacultura portuguesa e também são espécies com maior importância em termos ibéricos e aquacultura europeia. A legislação que regula a atividade da aquicultura, especialmente quando é desenvolvida em áreas do domínio público, é muito dispersa e complexa, em particular pelo grande número de entidades envolvidas. Um aspeto muito importante no processo de produção é a quantidade adequada de alimento, que é calculada em função da biomassa e da temperatura da água, a fim de otimizar os índices de crescimento, FCR, SGR, DGI e K. A aplicação de tecnologias de visão por computador na aquacultura é complicada, os peixes são sensíveis, suscetíveis a stress e livres para se deslocar num ambiente em que a iluminação, a visibilidade e a estabilidade não são controláveis na maioria dos casos, portanto o projeto Aquatropolis tem como objetivo principal desenvolver recursos tecnológicos para otimizar as operações, reduzir os riscos e as incertezas que afetam o processo produtivo e, dessa forma, garantir mais rendimento aos produtores, qualidade e segurança alimentar para o consumidor final.

A abordagem metodológica do presente estudo foi estruturada em 3 tarefas: sistematização do processo de licenciamento aquático e revisão da legislação portuguesa; descrição dos modelos de produção aplicados nas principais espécies produzidas em Portugal e validação experimental dos protocolos de crescimento.

A compilação do licenciamento e legislação permite ao produtor uma leitura melhor e um fácil acesso, sendo uma ótima ferramenta de suporte. Os modelos de produção foram projetados para a automação de operações através de ordens de produção feitas para intervenção humana ou de equipamentos, combinadas com os diferentes parâmetros hidrológicos para uma produção inteligente que pode ser adaptada ao melhor custo-benefício. Para a validação dos protocolos de crescimento, concluiu-se que o melhor método de alimentação é a tabela de alimentação mostrando um melhor FCR, com a ajuda da Hydrology DS5 Water Quality Multiprobe com medição constante de parâmetros hidrológicos.

**Palavra-chave:** Legislação, licenciamento, modelos de produção, parâmetros hidrológicos, *Sparus aurara*, *Dicentrarchus labrax*, novas tecnologias, Aquatropolis.

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## *Abstract*

Fisheries and aquaculture remain important sources of food, nutrition, income and livelihoods for hundreds of millions of people around the world. World per capita fish supply reached a new record high of 20 kg in 2014, thanks to vigorous growth in aquaculture, which now provides half of all fish for human consumption, and to a slight improvement in the state of certain fish stocks due to improved fisheries management. Presently, a quarter of seafood products consumed in the EU are produced on farms, in 2011, 1.24 million tonnes of aquaculture goods were produced in the EU, worth €3.51 billion. The production models have as objectives the development of strategies that help aquaculture become more efficient and less harmful to the environment, the species seabass and gilt-head bream are representative species of Portuguese aquaculture and also, they are species with greater importance in terms of Iberian and European aquaculture. Legislation regulating aquaculture activity, especially when it is developed in areas of the public domain is too dispersed and complex, in particular by the large number of entities involved. A very important aspect in the production process is the adequate dosage of feed, which is calculated as a function of biomass and water temperature, in order to optimize growth rates, FCR, SGR, DGI and K. Application of computer vision technologies in aquaculture is complicated, the inspected subjects are sensitive, susceptible to stress and free to move in an environment in which lighting, visibility and stability are not controllable in most cases so the Aquatropolis project has as main objective develop technological resources to optimize operations, reduce risks and uncertainties affecting the productive process, and in that way ensuring more income for producers, quality and food safety for the final consumer.

The methodological approach of the present study was structured on 3 tasks: systematization of the aquaculture licensing process and review of the Portuguese legislation; description of the production models applied on the main species produced in Portugal and experimental validation of growth protocols.

The licensing and legislation compilation allows the producer a better reading and easy access being a great tool of support to the producer. The production models were designed for the automation of operations through production orders made for human intervention or equipment operations, combined with the different hydrological parameters to an intelligent production that can be adapted to the best cost-benefit. For the validation of growth protocols, we conclude that the best feeding method is the feeding table showing a best FCR, with help of the Hydrolab DS5 Water Quality Multiprobe with constant measuring of hydrological parameters.

**Keywords:** Legislation, licensing, production models, hydrological parameters, *Sparus aurata*, *Dicentrarchus labrax*, new technologies, Aquatropolis.

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## *List of abbreviations*

| | |
|---|---|
| **AGR** | Absolute Growth Rate |
| **APA** | Aquaculture Production Area (Área de Produção Aquícola) |
| **BPAO** | Byproducts of Processed Animal Origin (SPOAT – Subprodutos de Origem Animal Transformados) |
| **CE** | Conformité Européenne |
| **CEE** | Comunidade Económica Europeia |
| **CGAP** | Code of Good Agricultural Practises (CBPA – Código de Boas Práticas Agrícolas) |
| **$CO_2$** | Carbon dioxide |
| **CRCD** | Commission for Regional Coordination and Development (CCDR – Comissão de Coordenação e Desenvolvimento Regional) |
| **DGI** | Daily Growth Index |
| **DGRM** | Directorate for Natural Resources, Safety and Maritime Services (Direção Geral de Recursos Naturais, Segurança e Serviços Marítimos) |
| **DO** | Dissolved oxygen |
| **DOC** | Dissolved organic carbon |
| **EIA** | Environmental Impact Asssessment (AIA – Avaliação do Impacte Ambiental) |
| **EU** | European Union |
| **FAO** | Food and Agriculture Organization |
| **FCR** | Food Convertion Ratio |
| **HRA** | Hydrographic Region Administration (ARH – Administração da Região Hidrográfica) |
| **I&D** | Investigation and Development |
| **INCB** | Institute for Nature Conservation and Biodiversity (ICNB – Instituto da Conservação da Natureza e da Biodiversidade) |
| **INE** | Instituto Nacional de Estatística (National Institute of Statistics) |
| **K** | Fulton Condition Index |
| **MSPP** | Maritime Space Planning Plan (POEM - Plano de Ordenamento do Espaço Marítimo) |
| **NER** | National Ecological Reserve (REN – Reserva Ecológica Nacional) |
| **PMTI** | Port and Maritime Transport Institute (IPTM – Instituto Portuário e dos Transportes Marítimos) |
| **RDAF** | Regional Directorates for Agriculture and Fisheries (DRAP – Direção Regional de Agricultura e Pescas) |
| **RDE** | Regional Directorates of Economy (DRE – Direções Regionais de Economia) |
| **RFID** | Radio frequency identification |

| | |
|---|---|
| **RGEL** | Regime Governing the Exercise of Livestock (REAP – Regime do Exercício da Atividade Pecuária) |
| **SGR** | Specific Growth Ratio |
| **SMEs** | Small and medium-sized enterprises |
| **T** | Temperature |
| **TGC** | Thermal Growth Coefficient |
| **UV** | Ultraviolet |
| **VCN** | Veterinary Control Number (NCV – Número de Controlo Veterinário) |
| **WSN** | Wireless sensor networks |

# 1. Introduction

## 1.1. Aquaculture in the world

Fisheries and aquaculture remain important sources of food, nutrition, income and livelihoods for hundreds of millions of people around the world. World per capita fish supply reached a new record high of 20 kg in 2014, thanks to vigorous growth in aquaculture, which now provides half of all fish for human consumption, and to a slight improvement in the state of certain fish stocks due to improved fisheries management. Recent reports by high-level experts, international organizations, industry and civil society representatives all highlight the tremendous potential of the oceans and inland waters now, and even more so in the future, to contribute significantly to food security and adequate nutrition for a global population expected to reach 9.7 billion by 2050 (FAO, 2016).

As stated in FAO, 2016 in 2014, fish harvested from aquaculture amounted to 73,8 million tonnes, with an estimated first-sale value of US$ 160,2 billion, consisting of 49.8 million tonnes of finfish (US$ 99.2 billion), 16.1 million tonnes of molluscs (US$ 19 billion), 6.9 million tonnes of crustaceans (US$ 36.2 billion), and 7.3 million tonnes of other aquatic animals including frogs (US$ 3.7 billion). Measured at the national level, 35 countries produced more farmed than wild-caught fish in 2014. Countries in this group include five major producers, namely, China, India, Viet Nam, Bangladesh, and Egypt. The other 30 countries in this group have relatively well-developed aquaculture sectors, e.g. Greece, the Czech Republic and Hungary in Europe, and the Lao People's Democratic Republic and Nepal in Asia.

According to FAO, 2016, by 2014, a total of 580 species and/or species groups farmed around the world, including those once farmed in the past, had been registered with production data by FAO. These species items include 362 finfishes (including hybrids), 104 molluscs, 62 crustaceans, 6 frogs and reptiles, 9 aquatic invertebrates, and 37 aquatic plants.

In agreement with FAO, 2016, many millions of people around the world find a source of income and livelihood in the fisheries and aquaculture sector. The most recent estimates indicate that 56.6 million people were engaged in the primary sector of capture fisheries and aquaculture in 2014. Of this total, 36 % were engaged full time, 23% part time, and the remainder were either occasional fishers or of unspecified status. For the first time since the period 2005–2010, the total engagement in fisheries and aquaculture did not increase. Overall employment in the sector decreased, almost entirely due to a decrease of about 1.5 million fishers, while engagement in aquaculture remained more stable. Consequently, the proportion of those employed in capture fisheries within the fisheries and aquaculture sector decreased from 83% in 1990 to 67% in 2014, while that of those employed in fish farming correspondingly increased from 17 to 33%.

## 1.2. Aquaculture in Europe

The aquaculture sector in Europe is diverse, encompassing traditional artisanal and family shellfish and pond-culture operations through medium-scale fish-farm businesses to multinational marine farming companies. The applied production technologies also show great diversity. However, more than 90 percent of the aquaculture farms are rather small farms that are geographically dispersed throughout Europe. The European aquaculture sector has emerged as an increasingly important contributor to the region's food production sector and has grown significantly over the last two decades (Varadi *et al.*, 2001).

Presently, a quarter of seafood products consumed in the EU (including imports) are produced on farms, in 2011, 1.24 million tonnes of aquaculture goods were produced in the EU, worth €3.51 billion. There are over 14 000 aquaculture enterprises in the EU, directly employing 85 000 people in total (Science for Environment Policy, 2015). Since European aquaculture is concentrated mainly in peripheral coastal and rural regions, there are also significant socio-economic benefits of aquaculture development in Europe. Much growth has been stimulated by consumer demand, as well as technological development work, making the different and various forms of production technically and economically viable (Varadi *et al.*, 2001).

The percentage contribution of European aquaculture to world aquaculture production has decreased since 1988 due to the higher rate of increase seen in other regions, however, it is growing in absolute terms (Figure 1).

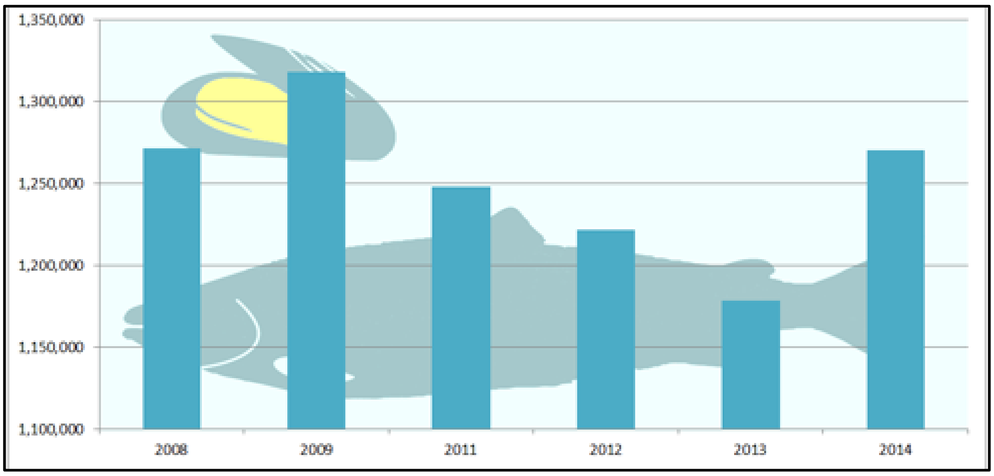


**Figure 1 -** Evolution of EU aquaculture production volume (2008 - 2014). Volume in tonnes live weight, EU - 28. [Source: EUROSTAT].

Although competition exists between wild and farmed supplies, the extent of direct substitution is variable and depends on the species concerned.

The contribution of aquaculture to food security has become and will remain important in certain regions of Europe, either directly, through consumption of the products, or indirectly, through the income and economic benefits obtained by the people involved in aquaculture and its related activities (Varadi *et al.,* 2001)

The main species produced in Europe are mussels, trout and salmon. Following with other species like oyster, seabream, carp and seabass (Figure 2).

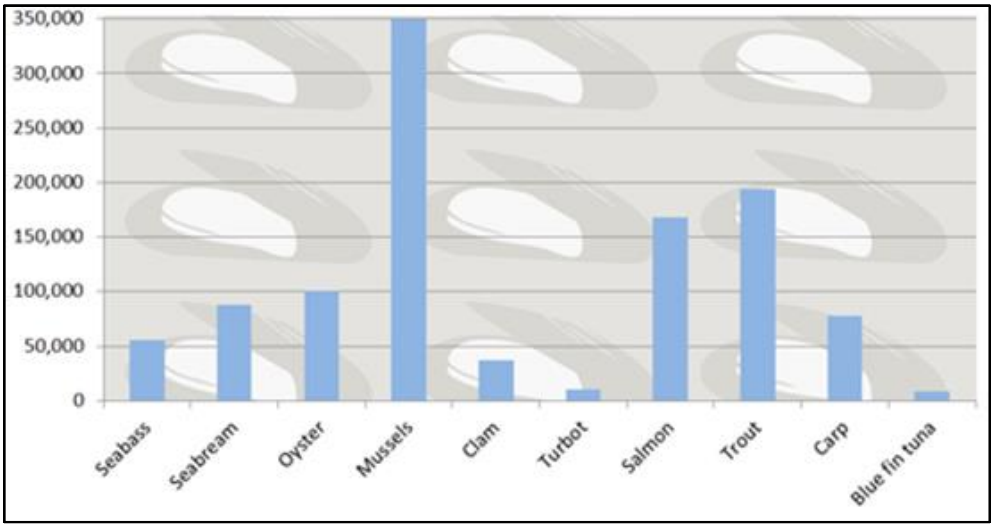


**Figure 2 -** Main aquaculture species in EU countries. Average volume in tonnes live weight 2008 - 2014. [Source: EUMOFA].

Europe produced 2 930,1 thousand tonnes in 2014 c0orresponding to a world percentage of 3.97%. Its main producers were Spain, United Kingdom, France, Italy and Greece (Figure 3).

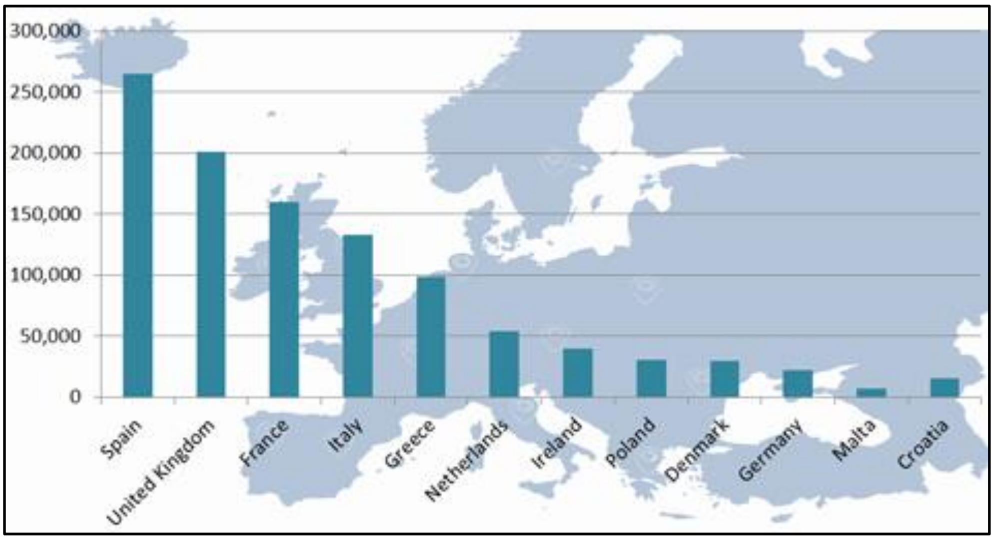


**Figure 3 - Main aquaculture producing EU countries. Average volume in tonnes live weight 2008 - 2015. [Source: EUROSTAT].**

In agreement with the European Commission, 2017, in Europe, aquaculture accounts for about 20% of fish production and directly employs some 85 000 people. The sector is mainly composed of SMEs or micro-enterprises in coastal and rural areas. EU aquaculture is renowned for its high quality, sustainability and consumer protection standards.

EU overall output has been more or less constant in volume since 2000 whereas global production, at the same time, has been growing by nearly 7% per year.

The Commission intends to boost the aquaculture sector through the Common Fisheries Policy reform, and in 2013 published Strategic Guidelines presenting common priorities and general objectives at EU level. Four priority areas were identified in consultation with all relevant stakeholders: reducing administrative burdens, improving access to space and water, increasing competitiveness and exploiting competitive advantages due to high quality, health and environmental standards.

On the basis of the guidelines, the Commission and EU countries are collaborating to help increase the sector's production and competitiveness. EU countries have been asked to set up multiannual plans to promote aquaculture. The Commission is helping with the identification of bottlenecks but also facilitates cooperation, coordination and exchange of best practices between EU countries.

### **1.3.** Aquaculture in Portugal

In Portugal, until the 1970s, aquaculture production was dominated by mugilids, typically forage species of low commercial value, which accounted for about 80% of fish production. The 1980s were characterized by a large increase in fish farms in inland waters (particularly rainbow trout), accompanied by bivalves (especially clams) in brackish and marine waters. The 1990s are characterized by the strong growth and modernization of aquaculture of marine species, initially focused on sea bass and gilt and, more recently, on turbot and sole (DGRM, 2014).

The use of salt industry infrastructures (yew, dikes, floodgates, monks, etc.) by marine aquaculture has emerged as a cheap and easy process to turn these neutralised facilities into productive units with lower environmental impacts. However, the size of the tanks, the characteristics of the bottoms and the water flow rates, are limiting factors of the productivity of this traditional aquaculture system, falling under the semi-intensive production regimes, termed "esteiro" or "estuarine" aquaculture. Still using traditional methodologies, in the areas between tides are bivalve nurseries, in extensive regime, that contribute with a very significant portion for the national aquaculture production. In recent years, some establishments have been installed on the coast, operating in an intensive regime, capturing sea water and producing, in particular, turbot and sole (DGRM, 2014).

Since the early 1990s, aquaculture production has increased from 4 457 tonnes in 1990, increasing its production in 2015 to 10 791 tonnes, amounting to 50 million euros, which is justified by the higher production of turbot and by its lower value in relation to the previous year, because of the increase of the production and the demand of fish of smaller sizes (DGRM, 2014; INE, 2016).

During this period, aquaculture production in fresh water has been reduced, both in absolute and relative terms, in relation to crops produced in marine or brackish waters, but increasing its production up to 2015 reaching 788 tonnes. The production in marine and brackish waters has therefore shown a growing trend, with the most commonly produced species being turbot, gilt-head bream/seabass, clams and oysters (DGRM, 2014). At the end of 2014, there were 1 521 licensed aquaculture establishments for fresh, salty and brackish water, 1 unit less than in 2013, with about 88,3% of ponds to produce bivalve molluscs, most of which are in Ria Formosa (Figure 4) (INE, 2016).

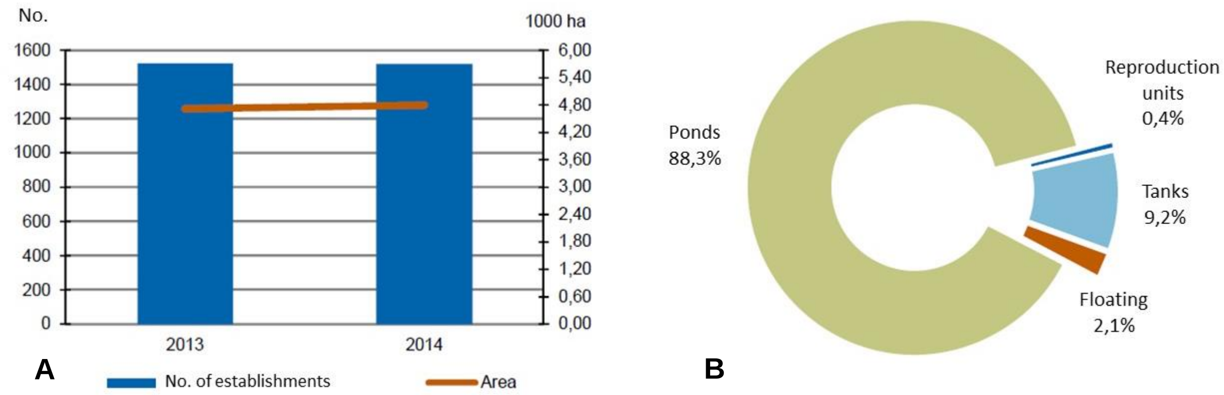


**Figure 4 - A: Number and area of aquaculture establishments in Portugal; B: Type of aquaculture establishments in Portugal. [Adapted from INE, 2016].**

Since the sector is constituted, in the overwhelming majority, by very small companies, with a few exceptions, many of the difficulties experienced may be minimized or even exceeded through a greater degree of associative, either through collaboration between companies, both with the manufacturing industry and also with the institutions of the scientific and technological system with a view to improve production processes.

There are three types of production in aquaculture: we have the extensive regime in which it exclusively takes advantage of the available natural conditions and where the control of the production is almost non-existent. In the semi-intensive regime, it still requires a low level of control, but there is some recourse to inert feed. In the intensive regime, there is full control of the entire production, high technology and only used inert food.

The production of aquaculture in fresh water is exclusively intensive. In salty and brackish water, the intensive regime, which strengthened its weight in 2014, it originated 39.2% of production, while the semi-intensive sector accounted for only 13.1% of total production (Figure 5). The decrease in semi-intensive production was due to the conversion of many fish farms to extensive bivalve production (DGRM, 2014).

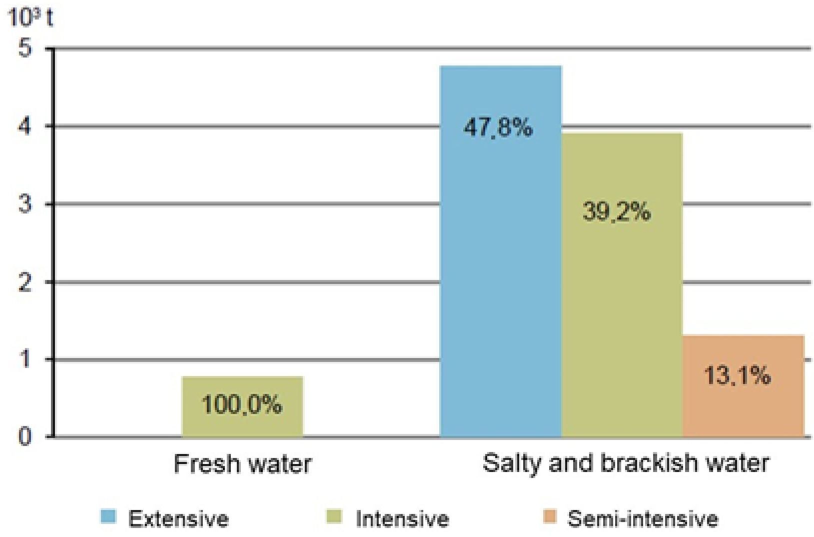


**Figure 5 - Aquaculture production by type of water and regime (2014). [Adapted from INE, 2016].**

The main species produced in Portugal in fresh water are rainbow and common trout; in brackish and marine water (fish, molluscs and crustaceans) are meagre (*Argyrosomus regius*), gilt-head bream (*Sparus aurata*), European eel (*Anguilla anguilla*), sole (*Solea solea*), turbot (*Psetta maxima*), seabass (*Dicentrarchus labrax*), white seabream (*Diplodus sargus*), clams (*Ruditapes decussatus*), common edible cockle (*Cerastoderma edule*), common periwinkle (*Littorina littorea*), ditch shrimp (*Palaemonetes varians*), grooved razor shell (*Solen marginatus*), mussels (*Mytilus edulis*), Japanese oyster (*Crassostrea gigas*) and Portuguese oyster (*Crassostrea angulata*) (Figure 6) (INE, 2016).

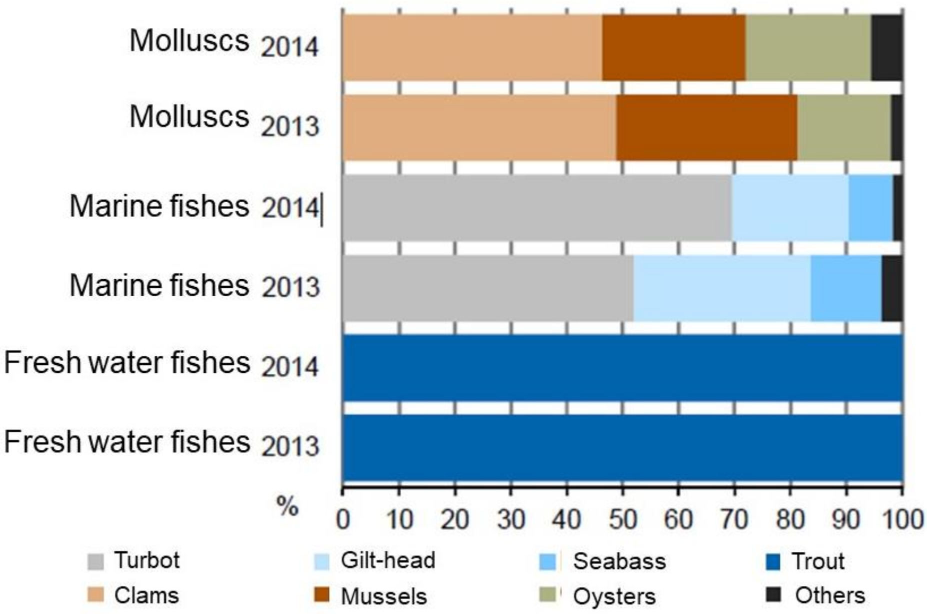


**Figure 6 - Structure of production volume in aquaculture by species (2013-2014). [Adapted from INE, 2016].**

### 1.3.1. Aquaculture representative species in the Portuguese context – Seabass and Gilt-head bream

Production protocols are the secret of any aquaculture. It is stealthy and only those who work in the company have access because there is the danger of plagiarism of other competing companies, information leakage by the workers and a poorly drawn protocol leads to losses. A well-crafted and optimized protocol will generate a greater profit margin. Protocols are associated with a species in the context of a production phase that have associated production models with the aim of planning and triggering production orders and other daily actions to be performed by people or equipment.

A production model is a set of tasks that should constitute the day-to-day of the professionals who operate the production. A model is defined as a schedule of activities, each of which will require action and the collection of data. The models have as objectives the development of strategies that help aquaculture become more efficient and less harmful to the environment, thus justifying the hope and investment put in this type of culture and the optimization of protocols for production of traditional species and development protocols for new species in maternity and organic production methods, allowing diversification of supply.

The species seabass and gilt-head bream are representative species of Portuguese aquaculture and also, they are species with greater importance in terms of Iberian and European aquaculture.

The gilt-head bream, *Sparus aurata*, is a bream belonging to the Sparidae family and is currently the most cultivated saltwater fish in the Mediterranean (Figure 7).

It is a fish compressed laterally with the convex head profile and well-armed with teeth. It is easy to distinguish by the characteristic yellow stripe on the forehead, between the eyes, and is most evident in adult individuals. Another characteristic is the presence of a black spot on the operculum with the beginning of the line. The head is large, with the profile of the convex face and small eyes. The muzzle and preopercle have no scales. The mouth is large, and the upper jaw extends beyond the middle of the eye. It presents some, very thick fleshy lips, the upper jaw and is slightly larger than the lower one. Both jaws have 4 to 6 canines located in the anterior part and then 2 to 4 sets of small but powerful molars capable of crushing shells of molluscs from which they feed. In the lower part of the operculum, which has a characteristic pink band that can sometimes spread to occupy most of the operculum. The body is oval, tall and long laterally. It has a continuous dorsal fin with 11 soft thorny rays and 13 below. The anal fin has 3 hard and 11-12 soft rays. The pectoral fins are long and pointed, reaching the back of the beginning of the anal fin. The caudal fin is bifurcated, with two pointed lobes. The body is covered with cycloid scales, between 75 and 85 on the lateral line, which is simple and continuous. The colour is darker in the back and quite light in the silvery grey of the belly. The dorsal fin is bluish grey, and has a medium black line, and the caudal fin is grey with black edges (Ortega, 2008).

**Figure 7 - *Sparus aurata*. [Source: Fishbase, 2017]**

It is common throughout the Mediterranean, it is also distributed along the eastern coast of the Mediterranean from Guinea and Senegal to the Bay of Biscay and the south of the British Isles. It is also present in the Canary Islands (Figure 8) (Sola *et al.,*2014).

**Figure 8 - Distribution of the Gilt-head bream. [Source: Fishbase, 2017]**

It is a marine species, very common in rocky and algae bottoms and *Posidonia oceanica*, which is not very often found on sandy or muddy bottoms. Young individuals have more coastal habits, living in bottoms of less than 30 meters, to which adults can reach 100-150 meters deep. Despite being a gregarious species, they often live alone or in small groups. However, reproductive migrations can form groups of thousands of individuals (Sola *et al.,*2014).

It is a eurytherm and euryhaline fish that is very sensitive to low temperatures, failing to feed if the temperature drops from 12-13 °C, which can withstand lower temperatures, being its minimum lethal in the order of 5-7 °C. On the contrary, it can withstand quite high temperatures, growing very fast at temperatures of 25-26 °C and withstand temperatures up to 32-33 °C. In relation to its euryhaline nature, its ecological plasticity is even greater: gilthead is a species that has been shown capable of living in salinity conditions varying between 3 and 70, crossing estuaries and coastal lagoons with a wide range of salinities (Ortega, 2008; Sola *et al*.,2014).

Their food is mostly carnivorous, preferring bivalve molluscs and small fish or cephalopods (Sola *et al.,*2014).

Its reproduction is hermaphrodite protandric type: first it behaves like male and later like females. During the first year of life they are immature, to which some can get to mature as males. But in most fish, the first sexual maturation only occurs during the second year of life. At this moment, the ventral part of the gonad develops, originating functional testicles. At the end of this reproductive phase, all individuals initiate a process of sexual inversion, which is completed by approximately 80% of the population that from the third year of life will behave like females. The remaining 20% interrupt reversion processes and remain male. However, this proportion also depends on other social factors. The maturation of the gonad is progressive, and this translates into a sequential set carried out for 2-4 months at a rate of about thousands of eggs per day. The total number of eggs per female is variable, ranging from 500 000 to 3 000 000 eggs/kg of females. The diameter of the eggs ranges from 0.9 to 1 mm. In nature, the posture season extends from November to February, which due to the posture zone may be delayed or delayed for a few weeks (Ortega, 2008; Sola *et al*.,2014).

The European seabass (Figure 9) is a percomorph of the Serranidae family. Its scientific name is *Dicentrarchus labrax* (Linnaeus, 1758) and is, together with gilt-head bream, the most important example of Mediterranean aquaculture, with a total production in 2015 exceeding 1 471 tonnes. There are other species of fish, some of which are freshwater, known as seabass: American seabass, *Morone saxatilis* (Walbaum, 1792), Asian seabass, *Lates calcarifer* (Bloch, 1790), etc. (Ortega, 2008).

Its body is broad and vigorous and is provided with large scales. The head is pointed, the mouth large, terminal and slightly protrusive. The nasal openings are relatively small, and the lower jaw is somewhat prominent. It has two dorsal fins, the first of which has 8-9 thorny rays and is triangular, while the second has 1 hard radius and 11-12 soft rays and is more irregular. The anal fin also has 3 hard spines rays, and the caudal fin is slightly bifurcated. The caudal peduncle is large, and the lateral line is complete, with 62-74 scales. Its colour is grey lead, darker in the dorsal part. The ventral part is yellowish and the sides are silver. These colours may vary depending on its area of distribution and the funds it is in. They have a diffuse black spot on the upper angle of the operculum. The pre-operculum has a serrated posterior margin and the operculum has two characteristic spines. The vomer has teeth, the shape of which is characteristic in *Dicentrarchus punctatus* (Bloch, 1792). Its maximum size can reach 80-100 cm, with sizes between 40 and 55 cm being more common. (With this maximum size, its weight oscillates between 2 and 5 kg). It does not present sexual dimorphism to which the females have the head wider and the body is higher than the males, they grow faster and reach larger size (Ortega, 2008; FAO, 2017).

**Figure 9 - *Dicentrarchus labrax*. [Source: Fishbase, 2017]**

In the Mediterranean, there are two species of the genus *Dicentrarchus*: *Dicentrarchus labrax* and *Dicentrarchus punctatus*. They differ fundamentally by the teeth of the vomer, which in *D. punctatus* has an arrowhead, while in *D. labrax* they adopt a V shape; while the eye of *D. labrax* is larger and only juveniles may have a dotted at the top, the eye of *D. punctatus* is smaller and has characteristic splashes on the flanks and back, it is permanent, even in adults. Another difference is that while *D. labrax* presents cycloidal scales on the upper part of the head, *D. punctatus* presents ctenoid scales. *D. labrax* reaches larger sizes, reaching almost 1 meter in length and weighing more than 10 kg. However, in *D. punctatus*, specimens weighing more than 1 kg are scarce (Ortega, 2008).

It is a specie of shallow coastal waters that only live grouped when they are young, but that only live alone or in couples when they are adults. They can be found on the rocky coasts and at the rivers mouths and salines, penetrating the estuaries and occasionally reassembling the rivers. It is a very voracious animal and feeds itself of small fishes and invertebrates of all type: squid, prawns, etc. (Ortega, 2008).

Its distribution area extends throughout the Mediterranean (Figure 10), including the Black Sea, and the East Atlantic stretches from the British Isles to the north, reaching as far as Norway and south to Morocco and the Canary Islands, reaching the coast of Senegal (FAO, 2017).

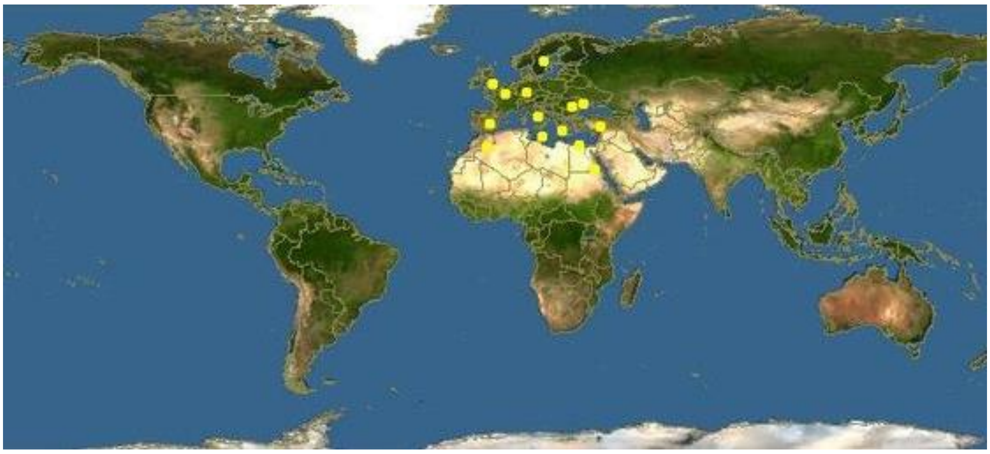

**Figure 10 - Distribution of the European seabass. [Source: Fishbase, 2017]**

It is a specie with great ecological plasticity, very euryhaline and eurythermal. Thus, they can live in almost sweet waters (salinity of 0.5) and withstand very high salinities, higher than 60. Although these changes are progressive, they have already been shown to be able to withstand abrupt changes of salinity in the order of 8-10. Regarding to temperature, they are capable of withstanding temperatures ranging from 3 to 30 °C. Its reproductive value is between 10 and 15 °C for Atlantic sea bass and 2 °C for Mediterranean seabass (Ortega, 2008; Eroldogan *et al.*, 2003).

Seabass is a very active predator (its name derives from the latin *lupus*, which means wolf), reason why its feeding is carnivorous. It feeds mainly on other fish and cephalopods, crayfish and less on other crustaceans such as crabs. In short, we can say that it is an opportunistic predator that feeds on everything that finds and that presents the appropriate size. Most of its movements, outside the breeding season, are related to the search for food, approaching coastal and estuarine environments. Below 10-12 °C their feed rate decreases considerably, so in winter they may leave the coastal lagoons returning to the open sea (Ortega, 2008; FAO, 2017).

It is a gonocoric specie that does not present sexual dimorphism. In the Mediterranean males generally reach sexual maturity from the second year of life, when they weigh more than 300-400g, while females do not do so until the third or fourth year of life when they weigh more than 500 -600g. However, in the Atlantic sea bass takes a year or two to reach its first sexual maturation. The seabass matures your gonad synchronously, and has only one posture in the year. The eggs are spherical and have 1 to 3 drops of fat that are fused to one after hatching. The size of the eggs according to the literature is between 1.1 and 1.5 mm in diameter, and the number oscillates around 300 000 eggs/kg of females. The variability in size is because there are differences according to the region; the eggs of Atlantic seabasses are larger in size (ranging from 1.2 to 1.5 mm), while eggs of Mediterranean seabass do not pass 1.2 mm in diameter. It is a fish with a winter stance; the natural time of laying goes from January to March, which may vary depending on the latitude: in

Mediterranean waters, can begin to reproduce if in December while in the Atlantic the posture can extend until May or June (Ortega, 2008; FAO, 2017).

### 1.4. Aquaculture legislation and licensing in Portugal

Legislation regulating aquaculture activity, especially when it is developed in areas of the public domain (about 90% of establishments) is too dispersed and complex, in particular by the large number of entities involved. This is accompanied by a series of administrative procedures that are too time-consuming and complex to obtain titles for the use and exploitation of water resources, allied until a few years ago, with too short deadlines for water use licenses (10/15 years) (DGRM, 2014).

The complexity of allocating new areas for aquaculture production, coupled with the need to involve various areas of knowledge, to which the economic agent must resort, makes the licensing process too costly and slow, a difficulty which is a barrier to entry. New investors that favour the renewal of the sector. The burden inherent in this difficulty may be reduced by allocating licences for the use of water resources for longer periods (DGRM, 2014). As Joe Healy, president of the Irish Farmers Association, "*many of the deficiencies and delays in licensing which has cost the industry millions of euro and hundreds of jobs.*"

However, some changes have been occurring since 2008, notably with the publication of Regulatory Decree No. 9/2008, of March 18, which allowed the creation of aquaculture production areas in the open sea. More recently, in the preparation of the Maritime Spatial Planning Plan (MSPP), other open sea areas have been identified that may be regulated for aquaculture purposes (DGRM, 2014).

### 1.5. Water quality – hydrological parameters

Successful aquaculture depends on providing animals with a satisfactory environment in which to grow. Good initial conditions for aquaculture can be assured by selecting a site with suitable soils and a high-quality water supply. An adequate environment must then be maintained over the culture period so that animals will survive and grow rapidly (Claude *et al*., 1998).

Water **temperature** is perhaps the most important variable affecting aquaculture production. Water temperature affects the natural productivity of aquatic ecosystems and directly or indirectly affects all other water quality variables. In aquaculture, it is seldom cost-effective to cool or heat large volumes of water, so the water temperature prevailing at a particular site determines which species can be cultured and the potential growth, health, and reproductive success of that species (Claude *et al*., 1998).

However, certain processes occur in narrow ranges of temperature, such as reproduction and growth. Out of this range fish are subjected to stressful conditions, which make them susceptible to diseases (manual del participante aquicultura, 2011). The requirement in temperature depends on the species of fish and the stage of development in this one is (egg, larva, post-larva or juvenile). On the other hand, there is an inverse relationship between the maximum amount of oxygen, which can dissolve in water and temperature. The higher the temperature, the lower the amount of oxygen in the water (Manual del participante aquicultura, 2011).

**Salinity** refers to the total concentrations of all ions in water. It is not, as some people think, the concentration of sodium chloride in water. The major ions contributing to salinity are calcium, magnesium, sodium, potassium, bicarbonate, chloride, and sulfate. The absolute and relative concentrations of these ions vary greatly among different waters. Each species of aquatic animal has an optimum range of salinity for reproduction and growth; outside that range, performance is diminished and survival may be poor. Fortunately, the salinity tolerance of most aquaculture species is rather wide and only large differences in salinity or sudden changes are likely to be important. Salinity also interacts with other water quality variables because the ionic strength of a solution affects equilibrium constants for all chemical reactions, and increasing the salt concentration of a solution decreases the solubility of dissolved gases through the "salting-out effect." Of practical importance, as the salinity of water increases, the solubility of dissolved oxygen decreases and the percentage of total ammonia present as toxic un-ionized ammonia decreases (Claude *et al*., 1998).

The **pH** value expresses the intensity of the acidic or basic character of water. It is defined as the negative logarithm of the hydrogen ion activity which, in fresh water, is essentially equal to the hydrogen ion concentration. The pH scale is usually represented as ranging from 0 to 14, but the pH can extend past those values. Conditions become more acid as pH values decrease and more basic as pH increases. Exposure of aquatic animals to extremes of pH can be stressful or lethal, but the indirect effects of pH and interactions of pH with other variables are usually more important in aquaculture than the direct toxic effects. Important interactions include the effects of pH on certain aqueous equilibria involving ammonia, hydrogen sulphide, chlorine, and metals. The fertility of aquatic ecosystems is also strongly influenced by environmental pH (Claude *et al*., 1998). Generally, pH values of 6.5 to 9 are most suitable for fish production. Extreme values impair growth and reproduction and can lead to death. The pH values may vary during the day depending on the photosynthetic activity and the concentrations of carbon dioxide in the water.

In addition to the above on pH, it is pertinent to point out that fish can be cultured in intervals of 6.5 to 9, and some can survive at more extreme pH. The changes of this parameter in a body of water are related to the concentration of carbon dioxide during photosynthesis, in such a way that this process determines in part the fluctuation of the pH, and this is how it rises during the day and decreases in the night (Manual de participante de aquicultura, 2011).

The availability of **dissolved oxygen** frequently limits the activities and growth of aquatic animals. Oxygen is the most important element in water for aquatic organisms, since animals need adequate quantities of this gas to perform the oxidative processes that allow them to obtain energy from the food (Manual de participante de aquicultura, 2011). Water saturated with oxygen contains 20-40 times less oxygen by volume than air and the energetic costs of breathing water are greater than for air because water is much denser and more viscous. The oxygen content in aquatic environments also varies much more than in air because the availability of oxygen is limited to begin with (oxygen is a sparingly soluble gas in water), so small differences in the metabolism of the aquatic community can dramatically change dissolved oxygen concentrations. If dissolved oxygen concentrations are consistently low, aquatic animals will not eat or grow well and will be susceptible to infectious diseases. If concentrations fall to very low levels, the animal may die (Claude *et al*., 1998).

**Ammonia** is the principal nitrogenous waste product excreted by crustaceans and most fishes. Some fish excrete significant quantities of urea, but it is rapidly hydrolysed in the environment to ammonia and carbon dioxide. Ammonia is also produced when nitrogen-containing organic matter decomposes. Accumulation of ammonia in aquaculture systems is undesirable because un-ionized ammonia is toxic to aquatic animals. Ammonia is also a source of combined inorganic nitrogen for plant growth and its availability may influence the productivity of aquatic ecosystems (Claude *et al*., 1998).

**Nitrite** ($NO_2^-$) is a naturally occurring intermediate product in two bacteria-mediated processes involving transformations of nitrogen in water and soils. Nitrite occasionally accumulates in aquaculture systems and can be toxic to aquatic animals. Nitrite is an intermediate in the process of nitrification, which is the two-step oxidation of ammonium to nitrate carried out by highly aerobic, gram-negative, chemoautotrophic bacteria. Nitrite normally does not accumulate in the environment because it usually is converted to nitrate as quickly as it is produced. Under certain conditions, however, the rate of ammonia oxidation can exceed the rate of nitrite oxidation and nitrite will accumulate. Nitrite is also an intermediate in denitrification, which is the biological reduction of nitrate to dinitrogen gas ($N_2$) or nitrous oxide ($N_2O$). Denitrification occurs under anaerobic conditions when heterotrophic bacteria use nitrate instead of oxygen as a terminal electron acceptor in respiration. Nitrite is an intermediate in the process and may accumulate in anaerobic soils and bottom muds (Claude *et al*., 1998).

**Turbidity** refers to an optical property of water that causes light to be scattered or absorbed rather than transmitted through the water in a straight line. Turbidity is caused by suspended material (such as soil particles, plankton, and organic detritus) and soluble coloured organic compounds. Turbidity caused by plankton generally is desirable in fish ponds. The relatively still, unmixed water in many aquaculture ponds favors sedimentation of solids, and suspended solids seldom exceed 100 or 200 mg/L for more than a few days. Even though turbidity caused by suspended soil particles will seldom have immediate direct effects on fish in ponds, in the long run it may harm fish populations. Clay turbidity will restrict light penetration, adversely affecting plant growth, and some of the particles will settle to the bottom and smother fish eggs and destroy benthic communities (Claude *et al*., 1998).

**Chlorophylls**, which are the primary photosynthetic pigments in phytoplankton, absorb light strongly in the blue and red wavelengths. Photosynthesis requires light and chlorophyll to convert carbon dioxide and water to sugar (carbohydrates), oxygen, and water. Chlorophyll allows photosynthesis. Plants use the sun energy stored in the carbohydrates through respiration. The respiration process employs carbohydrates and oxygen to produce carbon dioxide, water, and energy. Plants take advantage of this energy for growth and reproduction (Parker, 2012). The minimum light requirement for net phytoplankton growth varies from less than 5 to over 20 µE/m2/s, and depends on species, nutrient status, water temperature, and other factors. Models of phytoplankton growth can be used to estimate standing crops corresponding to maximum net primary production, but such estimates can vary widely depending on the expressions used to estimate gross photosynthesis and respiration. Results of simple empirical and mechanistic models under various simulation conditions indicate that values of 50-250 µg/L chlorophyll *a* appear to be reasonable approximations of the range of phytoplankton biomass over which net primary production is maximized. That range of values compares to values of 60-150 µg/L chlorophyll *a* cited by Boyd (1990) as typical of productive fertilized fish and shrimp ponds (Claude *et al*., 1998). Parker (2012) also confirms that unfertile ponds range up to 20 micrograms per liter (µg/l), and fertile ponds with rich phytoplankton blooms range from 20 to 150 µg/l.

Short-term changes in phytoplankton community organization in eutrophic ecosystems typically involve a fairly rapid (days to weeks) transition from a diverse community of moderate standing crop to a less diverse community of high standing crop. The resulting community, which is often dominated by only a few species, is, by nature, unstable because changes in the standing crop of those few dominant species dramatically affect overall phytoplankton community standing crop. Quite often the dominant phytoplankton in high-biomass communities are certain species of scum-forming **blue-green algae (cyanobacteria)** that are subject to sudden "die-offs" or "bloom collapses." In extreme instances, the entire phytoplankton population dies over a period of a day or two, and the combination of reduced photosynthesis and rapid decomposition of the dead algal cells may deplete the water of dissolved oxygen and cause fish kills. (Claude *et al*., 1998; Paerl *et al*., 1995).

Light, through its role in photosynthesis, is obviously an important factor affecting phytoplankton growth. The light energy needed for plant growth is commonly expressed as the intensity of light in the wavelengths that promote photosynthesis (generally considered to be in the range 400-700 nm). This is called **photosynthetically active radiation**, or **PAR**. The photosynthetic apparatus of phytoplankton is adapted to underwater conditions where light intensity is much lower than terrestrial conditions and, consequently, phytoplankton photosynthesis can be inhibited when light intensity is high. Phytoplankton photosynthesis may be partially inhibited when PAR exceeds 200-800 $\mu E/m^2/s$ and may become totally inhibited when PAR exceeds 1400 $\mu E/m^2/s$ (Claude *et al*., 1998; Aguirre-von-Wobeser *et al*., 2000).

**Biochemical oxygen demand (BOD)** is a standard test for organic material. This test is determined by measuring the dissolved oxygen in a freshly collected sample and comparing it to the dissolved oxygen level in a sample collected at the same time, but incubated at 20 °C for five days. The difference between the two oxygen levels is the BOD. On a per-hour basis, 0,5 mg/l would be considered rich (Parker, 2012).

Dissolved ionic substances can be measured by electrical conductance. On laboratory reports, this may be shown as specific **conductivity**. Conductivities in natural surface water measure from 50 to 1500 microohms per cm (Parker, 2012).

**Table 1 - Impact of hydrological parameters on aquaculture productivity.**

| Parameter | Impact on Aquaculture Productivity |
|---|---|
| **Temperature** | • Certain processes occur in narrow ranges of temperature, such as reproduction and growth.<br>• Out of the optimum range fish are subjected to stressful conditions, which make them susceptible to diseases.<br>• The requirement in temperature depends on the species of fish and the stage of development in this one is (egg, larva, post-larva or juvenile).<br>• There is an inverse relationship between the maximum amount of oxygen, the higher the temperature, the lower the amount. |
| **Salinity** | • Outside the optimum range for reproduction and growth, performance is diminished and survival may be poor.<br>• As the salinity of water increases, the solubility of dissolved oxygen decreases and the percentage of total ammonia present as toxic un-ionized ammonia decreases. |
| **pH** | • Exposure of aquatic animals to extremes of pH can be stressful or lethal, but the indirect effects of pH and interactions of pH with other variables are usually more important in aquaculture than the direct toxic effects.<br>• Extreme values impair growth and reproduction and can lead to death.<br>• The pH values may vary during the day depending on the photosynthetic activity and the concentrations of carbon dioxide in the water. |
| **Dissolved oxygen** | • Limits the activities and growth of aquatic animals.<br>• If dissolved oxygen concentrations are consistently low, aquatic animals will not eat or grow well and will be susceptible to infectious diseases.<br>• If concentrations fall to very low levels, can lead to death. |
| **Ammonia** | • Accumulation of ammonia is undesirable because un-ionized ammonia is toxic to aquatic animals. |
| **Nitrite ($NO_2^-$)** | • Occasionally accumulates in aquaculture systems and can be toxic to aquatic animals. |

| Parameter | Impact on Aquaculture Productivity |
|---|---|
| **Turbidity** | • Caused by plankton generally is desirable in fish ponds.<br>• Caused by suspended soil particles will seldom have immediate direct effects on fish in ponds, in the long run it may harm fish populations.<br>• Clay turbidity will restrict light penetration, adversely affecting plant growth.<br>• Some of the particles will settle to the bottom and smother fish eggs and destroy benthic communities. |
| **Chlorophyll** | • Chlorophyll allows photosynthesis, releasing oxygen into the water during day.<br>• At night consumes de oxygen leading to a drop level of oxygen in the tank. |
| **Blue-green algae (cyanobacteria)** | • Sudden "die-offs" or "bloom collapses." In extreme instances, the entire phytoplankton population dies over a period of a day or two.<br>• The combination of reduced photosynthesis and rapid decomposition of the dead algal cells may deplete the water of dissolved oxygen and cause fish kills. |
| **Photosynthetically Active Radiation (PAR)** | • Is an important factor affecting phytoplankton growth.<br>• Phytoplankton photosynthesis can be inhibited when light intensity is high, because the photosynthetic apparatus of phytoplankton is adapted to underwater conditions where light intensity is much lower than terrestrial conditions. |
| **Biochemical Oxygen Demand (BOD)** | • Used to assess the organic pollution in water systems. |
| **Conductivity** | • Low conductivity values may indicate high primary production, evidence of high food availability for fish or even when intense local eutrophication occurs.<br>• Is directly associated with salinization of the body of water in which low values of conductivity indicate low salinity. |

### **1.6.** Growth Rates

In the grow-out phase of an aquaculture, frequent sampling and calibration is performed to optimize fish growth (Diniz, 1998). Growth is directly related to environmental factors, such as temperature, salinity, dissolved oxygen, etc., and with food.

If the abiotic conditions are not suitable for the cultivated species, the animals will not grow as expected, there is a delay in their growth, as if it is fed incorrectly, with inadequate feed, their nutrition will not meet their needs changes in their normal growth. Thus, it is very important to evaluate the growth, since it indicates if the fish cultured is healthy or sick and if the food administered is correct for the species/stage.

A very important aspect in the production process is the adequate dosage of feed, which is calculated as a function of biomass and water temperature, in order to optimize growth rates.

Food Conversion Ratio (FCR) is defined as the ratio of body weight gain (g) per dry feed consumed (g), corresponds to the dry weight/wet weight produced, varying between 1.3 and 1.8 in optimal conditions for gilt-head seabream (Batista, 2008), and between 1.2 and 2.5 for sea bass (Ercan, *et al*., 2015).

The Specific Growth Ratio (SGR) is important in estimating and predicting the weight gain over a given period of time, under a defined temperature and conditions. This is the instantaneous growth rate calculated by logarithms of body weight (Houlihan, *et al*., 2001), corresponding to body weight variation over time (%).

The Daily Growth Index (DGI) is relatively similar to SGR, however, it assumes that the growth is proportional to the body mass in two thirds, allowing a better comparison between the growth rate of different groups, varying little independently of the weight (Guillaume, *et al.,* 1999).

Fulton Condition Index (K) is based on the weight-length relationship, is an indicator of the fish's physiological condition and was determined by the ratio (Cunha, *et al.,* 2016).

### **1.7.** New technologies in aquaculture

For the last three decades, computer vision technology is now a common sophisticated inspection technology. Advances in hardware have resulted in cameras and peripheral equipment with higher sensitivity and faster capabilities that are less expensive, and simpler to use and incorporate into control systems. Advances in image processing and classification methods have enabled the rapid extraction of fine details from images and more accurate data interpretation for control decisions. As a result, computer vision technologies are being used by almost all industries for a variety of inspection tasks. However, these technologies are still not widely used in aquaculture (Zion, 2012).

Application of computer vision technologies in aquaculture is complicated, the inspected subjects are sensitive, susceptible to stress and free to move in an environment in which lighting, visibility and stability are not controllable in most cases. The equipment must operate underwater or in a wet environment and is expected to be inexpensive (Zion, 2012). But these technologies are also important for all production systems: counting, size measurement and mass estimation, gender detection and quality inspection, species and stock identification, and the monitoring of welfare and behaviour. A few publications of work conducted in the context of fisheries, ecology or the postharvest industry are also described, in cases in which the inspection task is similar in these fields or where an image-processing algorithm or method could also be useful in aquaculture (Zion, 2012).

Other studies were conducted using all types of sensors, for example, DeCew *et al.* (2013) used acoustic sensors for field measurements of cage deformation, Simbeye *et al.* (2014) designed and implemented a wireless sensor network for tracking and control aquaculture based on virtual instruments and Zhuiykov (2011) did a solid-state sensors for on-line monitoring of such water quality parameters as pH, dissolved oxygen (DO), conductivity, turbidity, dissolved organic carbon (DOC) and dissolved metal ions. Other use of technologies is for traceability where Parreño-Marchante *et al.* (2013) presents a novel traceability system architecture based on web services, which are used to integrate traceability data captured through Radio Frequency Identification (RFID) systems with environmental data collected with Wireless Sensor Networks (WSN) infrastructure. The solution, suitable to be deployed in Small to Medium Enterprises (SMEs), is provided by integrating information collected along the entire food supply chain, tracking the products from the farm to the consumer. On this project is going to be used a probe connected to sensors that will transmit to *Fog computing* which is a hybrid solution with local intelligence and *cloud* processing capability, applicable in inshore and offshore realities and locations with poor connectivity. So, the *Fog Computing* expands the *Cloud Computing* paradigm to the network frontier.

### **1.8.** Aquatropolis project

This dissertation is part of the Aquatropolis project funding by COMPETE 2020, which is the new consortium that was created to promote the sustainable development of aquaculture. The consortium is formed by Compta, ALGAplus, Domatica, Polytechnic Institute of Tomar, Tagus Valley and the Polytechnic Institute of Leiria. The focus of the Aquatropolis project is to use of technology and industry 4.0 at the service of aquaculture. One of the main objectives of the project is to promote the democratization of state-of-the-art technologies in the sector, allowing most of the Iberian business community in aquaculture to have the technological resources to optimize operations, reduce risks and uncertainties affecting the productive process, and in that way ensuring more income for producers, quality and food safety for the final consumer.

**Figure 11 –Aquatropolis logo and with the respective cofinanciers.**

The Aquatropolis project will take advantage of the potential of the Internet of Things (IoT), “cloud” and “fog” computing systems, principles promoted by the "Sharing Economy" development model and finally technological innovation applied to precision instrumentation, guaranteeing scales that allow the introduction of leading-edge technologies in entrepreneurial initiatives with low investment capacity or emerging sustainable production models, such as multi-trophic aquaculture. Besides the high market potential, "Aquatropolis" will contribute to promote the productive efficiency, food quality and safety, better information for the consumer and, above all, the control of aquaculture activities in the maritime space, assuring the balance between social, economic and environmental perspectives.

The Aquatropolis framework born to empower the aquacultures with technological advances and predictive capacities to allow the intelligent control of the whole productive cycle, increased the efficiency of the production process and the effective use of resources.

**Objectives:** This dissertation aims to compile the most important licensing and legislation serving as a support tool to the fish farmer, the construction of production models to apply in electronic platforms designed to support producers in their daily activities and validation of growth protocols for *Sparus aurata* and *Dicentrarchus labrax* with intensive monitoring of water quality parameters using "fog" computing systems.

# 2. Material and Methods

The methodological approach of the present study was structured on 3 tasks: (1) systematization of the aquaculture licensing process and review of the Portuguese legislation; (2) description of the production models applied on the main species produced in Portugal and (3) experimental validation of growth protocols. All the data obtained on the present study will be used by the Aquatropolis consortium for the definition of the technological solution.

## 2.1. Licensing and legislation

For the accomplishment of this task it was necessary an intensive research of legislation and licensing about the sector.

From the research on the legislation and licensing of the sector a table with the important legislation and referring to the aquaculture sector was constructed, providing a greater facility of consultation by the producer.

## 2.2. Production Models

For the accomplishment of this work it was necessary an intensive bibliographical research.

With this burden of information and with the aim of clarifying and understanding some issues, a questionnaire was applied to fish farming's with semi-intensive regime.

This questionnaire (Annex I) was then applied in several aquacultures throughout the country, aiming to have a perspective of the sector under study at the national level.

With all the information collected it was then possible to build a production model to each species (gilt-head bream and seabass).

## 2.3. Validation of growth protocols

For the accomplishment of this work it was necessary an intensive bibliographical research about the hydrological parameters about the species under study.

### 2.3.1. Hydrological parameters

With the questionnaire (Annex I) complete and then made in some aquaculture companies, it was realized that only a few parameters were important to make their measurement, therefore only those were considered.

The following table has been drawn,

**Table 2 - Hydrological parameters for adults of gilt-head bream and seabass.**

| Hydrological Parameter | ***Sparus aurata* & *Dicentrarchus labrax* (Adult)** | | |
|---|---|---|---|
| | **Minimum** | **Optimum** | **Maximum** |
| **Dissolved oxygen (saturation)** | 80% | 90% | 100% |
| **Dissolved oxygen** | 5 mg/L | 6 mg/L | Saturation |
| **pH** | 7.5 | 7.7 – 7.8 | 8.5 |
| **Temperature** | 10 °C | 20 - 22 °C | 32 °C |
| **Salinity** | 30 | 35 - 37 | 40 |

#### 2.3.2. Sampling

Three trials were carried out with a duration of one month each in August, September and October. At the beginning and end of each trial all the individuals were measured and weighed in a precision scale (Adam PGL 3002), these measurements were made with the objective of each measurement terms the calculation of the biomass of the tank. Prior to sampling the specimens were anesthetized with 2 – phenoxietanol (VWR) (0.5 ml/L).

#### 2.3.3. System and Feeding

The food given was Standard Orange 4 provided by the feed company Sorgal- Sociedade de Óleos e Rações S.A.

In a recirculating system with 1600 L (Figure 12), consisting of 1 tank (with a sump, UV, recirculation pump, bag filters (200 µm and bio-balls) with 32 individuals (7 *Dicentrarchus labrax* and 25 *Sparus aurata*), on the first two trails the parameters (Table 2) were measured with the assistance of a YSI Professional Series probe (Figure 13) half hour prior to feeding, during feeding and half an hour after feeding.

**Figure 12 -** Experimental tank.


**Figure 13 -** YSI Professional Series probe**.**

On the first trial, the food was given two times a day and following a feeding chart (Figure 14). The feeding chart allows us to calculate the daily feed dose, relating mean weight to water temperature.

| | T (°C) | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Pi (g) | 10 | 12 | 14 | 16 | 18 | 20 | 22 | 24 | 26 | 28 |
| 10 | 0,4% | 0,7% | 1,0% | 1,3% | 1,6% | 1,9% | 2,1% | 2,4% | 2,6% | 2,9% |
| 25 | 0,4% | 0,6% | 0,9% | 1,1% | 1,3% | 1,6% | 1,8% | 2,0% | 2,2% | 2,4% |
| 50 | 0,3% | 0,5% | 0,8% | 1,0% | 1,2% | 1,4% | 1,6% | 1,8% | 1,9% | 2,1% |
| 75 | 0,3% | 0,5% | 0,7% | 0,9% | 1,1% | 1,3% | 1,5% | 1,6% | 1,8% | 2,0% |
| 100 | 0,3% | 0,5% | 0,7% | 0,9% | 1,0% | 1,2% | 1,4% | 1,6% | 1,7% | 1,9% |
| 125 | 0,3% | 0,5% | 0,6% | 0,8% | 1,0% | 1,2% | 1,3% | 1,5% | 1,6% | 1,8% |
| 150 | 0,3% | 0,5% | 0,6% | 0,8% | 0,9% | 1,1% | 1,2% | 1,4% | 1,5% | 1,6% |
| 175 | 0,3% | 0,4% | 0,6% | 0,8% | 0,9% | 1,0% | 1,2% | 1,3% | 1,4% | 1,6% |
| 200 | 0,3% | 0,4% | 0,6% | 0,7% | 0,9% | 1,0% | 1,1% | 1,2% | 1,4% | 1,5% |
| 225 | 0,3% | 0,4% | 0,6% | 0,7% | 0,8% | 1,0% | 1,1% | 1,2% | 1,3% | 1,4% |
| 250 | 0,3% | 0,4% | 0,5% | 0,7% | 0,8% | 0,9% | 1,0% | 1,2% | 1,3% | 1,4% |
| 275 | 0,3% | 0,4% | 0,5% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% | 1,3% |
| 300 | 0,3% | 0,4% | 0,5% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% | 1,3% |
| 325 | 0,3% | 0,4% | 0,5% | 0,6% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% | 1,2% |
| 350 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% |
| 375 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% |
| 400 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,2% |
| 425 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% | 1,1% |
| 450 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,0% | 1,1% |
| 475 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 0,9% | 1,0% | 1,1% |
| 500 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,8% | 0,9% | 1,0% | 1,1% |
| 525 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,7% | 0,8% | 0,9% | 1,0% | 1,1% |
| 550 | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,7% | 0,8% | 0,9% | 1,0% | 1,0% |
| 575 | 0,3% | 0,4% | 0,5% | 0,6% | 0,6% | 0,7% | 0,8% | 0,9% | 1,0% | 1,0% |
| 600 | 0,3% | 0,4% | 0,5% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 0,9% | 1,0% |
| 625 | 0,2% | 0,4% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 0,9% | 1,0% |
| 650 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,9% | 0,9% | 1,0% |
| 675 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,8% | 0,9% | 1,0% |
| 700 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,8% | 0,9% | 1,0% |
| 725 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,8% | 0,9% | 1,0% |
| 750 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,8% | 0,8% | 0,9% | 0,9% |
| 775 | 0,2% | 0,3% | 0,4% | 0,5% | 0,6% | 0,7% | 0,7% | 0,8% | 0,9% | 0,9% |

**Figure 14 -** Feeding chart supplied by the company Sorgal- Sociedade de Óleos e Rações S.A. for *Sparus aurata.*

On the second trial, the food was given three times a day and given until the fish were satiated.

On the third trial, the amount of food given in the first two trials was calculated and the cost of feed was calculated, also taking into account the growth indexes, the best feeding method (satiety or feeding chart) was chosen.

The method chosen was the feeding chart and in this test the parameters were measured with the Hydrolab DS5 Water Quality Multiprobe (Figure 15) that measured the parameters second to second for the "*fog*".

In this trial the food was given two times a day.


**Figure 15 -** Hydrolab DS5 Water Quality Multiprobe.

2.3.4. <u>Growth Rates</u>

With the data provided by the three trials, several growth rates were calculated:

- **Food Conversion Ratio (FCR)**

$$FCR = \frac{Total\ food\ given\ (g)}{Weight\ gain\ (g)}$$

- **Specific Growth Rate (SGR)**

$$SGR\ (\%/day) = \frac{\ln(final\ weight) - \ln\ (initial\ weight)}{Time} \times 100$$

- **Fulton Condition Index (K)**

$$K\ (\%) = \frac{Weight\ (g)}{Lenght^3 (cm)} \times 100$$

- **Daily Growth Index (DGI)**

$$DGI\ (\%) = \frac{Final\ weight^{\frac{1}{3}} - Initial\ weight^{\frac{1}{3}}}{Time\ (days)} \times 100$$

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# 3. Results

## 3.1. Licensing and legislation

After the information gathered from the research and the questionnaires made, all the important licensing and legislation were collected.

This licensing and legislation compilation allows the producer a better reading and easy access being a great tool of support to the producer.

### 3.1.1. Licensing Entities

Licensing of marine aquaculture activity is given by the Directorate General of Natural Resources, Safety and Maritime Services (DGRM), Regulatory Decree 14/2000 of September 21 as coordinating entity.

**HRA - Hydrographic Region Administration and other entities that license the use of water resources (ARH – Administração da Região Hidrográfica)**

Decree-Law no. 226-A/2007, of May 31, the issue of the titles of use of water resources, which necessarily precede the licensing of the activity of marine culture establishments, is the responsibility of the territorially competent Hydrographic Region Administration, called HRA.

In the areas of the public water domain affecting the port entities, HRA's powers to license and inspect water resources, from the date of entry into force of the ordinances referred to in no. 1 of the 13$^{th}$ article of the Water Law, approved by Law no. 58/2005, of December 29, are considered to be delegated to the Port Administration with jurisdiction in the locality, and the provisions of article 38 of Decree-Law no. 226-A/2007, of May 31, and in paragraph no. 1 of the 32$^{th}$ article of Decree-Law no. 97/2008, of June 11.

Provision is also made for the possibility of the HRA delegating its powers in licensing and monitoring of water resources to other entities, namely in the Institute for Nature Conservation and Biodiversity (INCB; ICNB – Instituto da Conservação da Natureza e da Biodiversidade), in accordance with Article 13 of Decree-Law no. 226-A/2007, of May 31, in conjunction with paragraph 7 of 9$^{th}$ article of Law no. 58/2005, of December 29.

When the issuance of titles of use of the public water domain may affect maritime safety, the preservation of the marine environment or other attributes of the Maritime Authority or port and shipping security, the Maritime Authority and PMTI (IPTM – Instituto Portuário e dos Transportes Marítimos). Are heard, respectively, by HRA, under the conditions set out in points f) and g) of paragraph 1 of 15$^{th}$ article of Decree-Law no. 226-A/2007, of May 31.

**DGRM- Directorate General of Natural Resources, Safety and Maritime Services (Direção Geral de Recursos Naturais, Segurança e Serviços Marítimos) /RDAF - Regional Directorates for Agriculture and Fisheries (DRAP – Direcções Regionais de Agricultura e Pescas)**

The Directorate General of Natural Resources, Safety and Maritime Services shall authorize the establishment of marine culture establishments and license their exploitation, in accordance with the provisions of articles 11 and 12 of Decree-Law no. 278/87, of July 7, with the word given by Decree-Law no. 383/98, of November 27, in conjunction with Regulatory Decrees no. 14/2000, of September 21 and with Regulatory Decree no. 9/2008, of March 18.

In the case of establishments to be located offshore and entering the Aquaculture Production Area, the installation authorization procedure begins with the application for allocation of title for use of water resources, to be issued by the competent HRA, With the prior opinion of the DGRM. When issued, the title of use of water resources (license) replaces the order of authorization of installation of the Director General of Fisheries and Aquaculture, and the operating license issued after the completion and approval of the installed unit.

**EIA Procedure (AIA – Avaliação do Impacto Ambiental)**

In relation to establishments under intensive production, there may be an Environmental Impact Assessment (EIA) procedure, in accordance with annex II of Decree-Law no. 69/2000, of May 3, with the changes introduced by Decree-Law no. 197/2005, of November 8, which is always prior to any licensing.

Intensive fish farming shall be subject to EIA where it is in a sensitive area or where the following conditions are met:

Fish farming in estuarine or similar systems or lagoon systems:

a) Tanks: area ≥ 5 ha or production ≥ 200 t/year or area ≥ 2 ha or production ≥ 80 t/year if, together with similar pre-existing units, less than 1 km away, the area ≥ 5 ha or production ≥ 200 t/year;
b) Floating structures: production ≥ 200 t/year or production ≥ 80 t/year if, together with similar pre-existing units, less than 1 km from each other, production results ≥ 200 t/year.

Marine fish farming: production ≥ 1000 t / year:

If there is an insolvency proceeding for the issue of the title of use of water resources (license), the EIA procedure will only begin after selection of the candidate to whom the title will be awarded.

**CRCD - Commission for Regional Coordination and Development (CCDR – Comissão de Coordenação e Desenvolvimento Regional)**

The installations to be in an area integrated in the National Ecological Reserve (NER; REN – Reserva Ecológica Nacional), require the prior authorization of the territorially competent CRCD, under the terms established in Decree-Law no. 166/2008, of August 22.

The necessary instructional elements in NER's authorization procedures are listed in annex II of Ordinance no. 1356/2008 of 28 November.

Establishments to be in protected areas and areas of the Natura 2000 Network, i.e. classified areas as defined in the legal regime for nature conservation and biodiversity (Decree-Law no. 142/2008, of 24 July), are also previously subject to the conditions of the applicable legislation, including the opinion of INCB and the submission to an Environmental Incidence Assessment procedure, when, in general terms, they are not subject to an EIA procedure.

### 3.1.1. Licensing

The table 3 describes the important licensing for aquaculture and is structured with the important procedures to open an aquaculture with its respective diploma and licensing entity.

**Table 3 - Important licensing for aquaculture.**

| Area | Diploma | Description | Entity |
|---|---|---|---|
| **Entity coordinating the Aquaculture sector** | Regulatory decree no. 14/2000, of September 21 | The licensing of marine aquaculture activity by the Directorate General for Maritime Resources (DGRM). | DGRM |
| | Decree-Law no. 278/87, of July 7 | The purpose of this decree-law is to define the legal framework for the practice of sea fishing and for the cultivation of marine species. | |
| | Decree-Law no. 383/98, of November 27 | Modified the Decree-Law no. 278/87, of July 7, about offenses in fisheries and marine cultures. | |
| | Regulatory decree no. 14/2000, of September 21 | It lays down requirements and conditions relating to the installation and operation of marine and related crop establishments, the allocation of authorizations and licenses and the conditions for their transmission and cessation. | |
| | Regulatory decree no. 9/2008, of March 18 | It defines the rules for the establishment of aquaculture production areas (APA) in the open sea and establishes the pilot aquaculture production area of Armona. | |
| **Use of water resources** | Decree-Law no. 226-A/2007, of May 31 | The issuance of titles of use of water resources, which necessarily precede the licensing of the activity of marine culture establishments, is the responsibility of the Hydrographic Region Administration, referred to as HRA. | HRA |
| | Decree-Law no. 97/2008, of June 11 | Establishes the economic and financial regime of water resources. | |

| Area | Diploma | Description | Entity |
|---|---|---|---|
| | Law no. 58/2005, of December 29 | Modified by the Decree-Law no. 245/2009, of September 22, Decree-Law no. 26/2010, of March 30 and Decree-Law no. 130/2012, of June 22.<br>Approves the Water Law by transposing Directive no. 2000/60/CE of the European Parliament and of the Council of 23 October into the national legal order. Establishes the foundations and institutional framework for sustainable water management. | |
| **Establishments under intensive production** | Decree-Law no. 69/2000, of May 3 | Establishes the legal framework for environmental impact assessment. | EIA |
| | Decree-Law no. 197/2005, of November 8 | The present diploma changes the Decree-Law no. 69/2000, of May 3, partially transposing into the internal legal order Directive no. 2003/35/CE of the European Parliament and of the Council of 26 May on public participation in the drafting of certain plans and programs relating to the environment, in so far as it amends Directive no. 85/337/CEE of the Council of 27 June, as amended by the Directive no. 97/11/CE of the Council of 3 March. | |
| **Facilities to be in an area integrated in the National Ecological Reserve (NER)** | Decree-Law no. 166/2008, of august 22 | Establishes the legal regime of the National Ecological Reserve (NER). | CRCD |
| | Annex II of the Ordinance no. 1356/2008, of November 28 | Establishes the conditions for the feasibility of the uses and actions referred to in numbers 2 and 3 of the 20th of Decree-Law no. 166/2008, of August 22. | |
| **Establishments to be in protected areas and areas of the Natura 2000 network** | Decree-Law no. 142/2008, of July 24 | Establishes the legal regime for the conservation of nature and biodiversity. | INCB |
| **Title of use of water resources** | Decree-Law no. 54/2005, of November 15 | Amended by the Law no. 78/2013, of November 21.<br>Establishes ownership of water resources. Repeals articles 1 of Decree no. 5787-III, of May 18, 1919, and Chapters I and II of Decree-Law no. 468/71 of November 5. | HRA |

#### 3.1.1.1. Other licenses

The table 4 describes other important licenses, authorizations and registrations for aquaculture that establishment of institutions may require the obtaining and his respective licensing entity and diploma.

**Table 4 - Other important licenses/authorizations/registrations for aquaculture.**

| | **Legal framework** | **Licensing entity** |
|---|---|---|
| **Works Licences Building permit; Authorization for use.** | Decree-Law no. 555/99, of December 16, modified by the Decree-Law no. 26/2010, of March 30. | City Council |
| **Approval of the Electrical Installation Project** | Legal background: Decree-Law no. 517/80 of October 31, modified by the Decree-Law no. 272/92 of December 3, and by the Decree-Law no. 315/95 of November 28; Decree-Law no. 26852, of July 30 of 1936, modified by the Decree-Law no. 446/76 of June 5. | Directorate-General for Energy and Geology and other entities included in these regulations. |
| **Register of Receiving Operator to Purchase Ration** | Decree-Law no. 245/99, of June 28. | Directorate-General for Veterinary Medicine |
| **Installation, Operation, Repair and Alteration of Equipment Under Pressure (Oxygen)** | Decree-Law no. 97/2000, May 25. | Regional Directorates of Economics (RDE), territorially competent |
| **Oil Warehouse Storage** | Decree-Law no. 389/2007, of November 30, conjugated with the Decree-Law no. 125/97, of May 13. | Local Councils or Regional Economic Directorates, territorially competent, depending on the intended storage capacity. |
| **Industrial Licensing of Establishments for Processing, Preparation and/or Production of Aquaculture Products** | Decree-Law no. 209/2008, of October 29. | RDAF, or Municipal Councils, depending on the type of establishment, assigning the Veterinary Directorate-General the Veterinary Control number to the establishments. |

#### 3.1.1.1. Activity Support Boats

Holders of marine culture establishments may be authorized to have boats to support the activity, which must be registered in the local auxiliary class. These vessels are used exclusively for the transport of crop products, as well as personnel, equipment and materials for the holding.

### 3.1.2. Legislation

The table 5 describes important legislation for aquaculture, it's systematized by areas and with its respective diploma for register of livestock receiving operator, veterinary products, veterinary medicines, animal by-products not intended for human consumption, traceability, labelling, nutrition labelling and waste and heavy metals.

**Table 5 - Important legislation for aquaculture.**

| Area | Diploma | Description |
|---|---|---|
| **Register of livestock receiving operator** | Ordinance no. 576/93, of June 4 | Considering the Decree-Law no. 1110/93, of April 10, which transposes into national law the Directive no. 89/662/CEE, of the Commission, of December 11, concerning veterinary checks on intra-community trade in animal products with a view to the completion of the internal market. |
| | Ordinance no. 100/96, of April 1 | In accordance with subparagraph a) of the no. 6 of the 5$^{th}$ article of the Regulation of Veterinary Controls Applicable to Intra-Community Trade in Products of Animal Origin, approved by the Ordinance no. 576/93, of June 4, traders to whom products of another Member State are supplied or who complete a complete batch of such products shall be subject to prior registration. |
| | Decree-Law no. 111/2006, of June 9 | The present Decree-Law transposes into national law the Directive no. 2004/41/CE, of the European Parliament and the Council, of April 21, which repeals the legislation on the hygiene of foodstuffs and the rules applicable to the production and marketing of certain products of animal origin intended for human consumption, and amending Directives no. 89/662/CEE and 92/118/CEE, of the Council, and the Decision no. 95/408/CE, of the Council. |
| | Decree-Law no. 152/2009, of July 2 | The present Decree-Law transposes into national law the Directive no. 2006/88/CE, of October 24, relative on animal health requirements for aquaculture animals and products thereof, and on the prevention and control of certain diseases in aquatic animals, as amended by Directive 2008/53/CE, of the Commission, of April 30, concerning spring viraemia of the carp. |

| Area | Diploma | Description |
|---|---|---|
| **Register of livestock receiving operator** | Decree-Law no. 169/1986, of June 27 | The present Decree-Law transposes for regulating the exercise of the activity of acquisition for resale of live animals; And it is advisable to institute a process leading to the establishment of a prior licensing, centrally organized and coordinated by the state departments, of the activity related to the acquisition for resale of live animals. |
| | Decree-Law no. 131/2008, of July 21 | The present Decree-Law transposes into national law the Directive no. 2007/10/CE, of the Commission, of February 21, which alters the annex II of the Directive no. 92/119/CEE, of the Council, of December 17, laying down general measures for the control of certain animal diseases and specific measures relating to swine vesicular disease. |
| **Veterinary products** | Decree-Law no. 237/2009, of September 15 | The present Decree-Law lays down the rules governing the manufacture, marketing, import, export, marketing and advertising of products for veterinary use. |
| | Order 460/2013, of January 9 | The Decree-Law no. 148/2008, of July 29, with the word given to it by Decree-Law no. 314/2009, of October 28, establishing a community code relating to veterinary medicinal products, regulates veterinary medicinal products, in particular those which were evaluated for the purpose of placing on the market as veterinary products, in particular veterinary medicinal products intended for minor species of company, and others, depending of their composition and indications for the target species, were classified as veterinary products. |
| **Veterinary medicines** | Decree-Law no. 148/2008, of July 29 | The present Decree-Law transposes, partially, into national law the Directives no. 2001/82/CE, of the European Parliament and of the Council, of November 6, establishing a community code relating to veterinary medicinal products, 2004/28/CE , of the European Parliament and of the Council, of March 31, which changes the Directive no. 2001/82/CE, establishing a community code relating to veterinary medicinal products, and 2006/130/CE, of the Commission, of December 11, which lays down criteria for exemption from the veterinary prescription for certain veterinary medicinal products applicable to food producing animals for human consumption. |

| Area | Diploma | Description |
|---|---|---|
| **Veterinary medicines** | Decree-Law no. 148/2008, of July 29 (Continuation) | The present Decree-Law transposes, equally, the Directive no. 91/412/CEE, of the Commission, of July 23, which establishes principles and guidelines of good manufacturing practices.<br>The present Decree-Law is also intended to ensure the implementation and ensure compliance in the national legal order with the obligations arising from Regulation (CE) no. 1084/2003, of the Commission, of June 3, concerning the examination of the variation of the terms of the marketing authorizations granted by the competent authorities of the Member States for medicinal products for human use and veterinary medicinal products in respect of veterinary medicinal products. |
| **Veterinary medicines** | Decree-Law 314/2009, of October 28 | The present Decree-Law transposes into national law the Directive no. 2009/9/CE, of the Commission, of February 10, which changes the Directive no. 2001/82/CE, establishing a community code relating to veterinary medicinal products. |
| | Ordinance 124/99, of February 17 | The present diploma lays down the standards to be met by clinical trials to be carried out on animals to ensure their physical integrity and the efficacy and safety of veterinary medicinal products. |
| | Ordinance 1048/2008, of September 16 | Approves the standards of good manufacturing practices for veterinary medicinal products. |
| | Ordinance 1049/2008, of September 16 | Approves the rules of good practice for the distribution of veterinary medicines. |
| | Order 3277/2009, of January 26 | It is therefore of the most importance to supplement the existing rules on veterinary medicinal products with effective means of monitoring their use at the level of livestock farms. In pursuit of this objective, the Decree-Law no. 148/2008, of July 29, requires the keeper of farm animals to keep up-to-date a register of veterinary medicinal products and veterinary medicines used on those animals. |
| | Order 460/2013, of January 9 | The Decree-Law no. 148/2008, of July 29, with the word given to it by Decree-Law no. 314/2009, of October 28, establishing a community code relating to veterinary medicinal products, regulates veterinary medicinal products, in particular those which were evaluated for the purpose of placing on the market as veterinary products, in particular veterinary medicinal products intended for minor species of company, and others that, depending their composition and indications for the target species, were classified as veterinary products. |

| Area | Diploma | Description |
| --- | --- | --- |
| **Animal by-products not intended for human consumption** | Regulation 1069/2009, of October 21 | It lays down health rules concerning animal by-products and derived products not intended for human consumption and repealing Regulation (CE) no. 1774/2002 (regulation on animal by-products). |
| | Decree-Law 122/2006, of June 27 | The present Decree-Law is intended to ensure the implementation and ensure compliance in the national legal order with the obligations arising from Regulation (CE) no. 1774/2002, of the European Parliament and of the Council, of October 3, laying down health rules concerning animal by -products not intended for human consumption, after referred to as Regulation. |
| | Ordinance 631/2009, of June 9 | The present Ordinance establishes the regulatory standards governing the management of effluents from livestock activities and the technical standards to be observed in the scope of the licensing of agricultural valorisation activities or the transformation of livestock effluents with a view to promoting adequate conditions of production, collection, storage, transport, appreciation, processing, treatment and final destination.<br><br>The present Ordinance establishes, still, Regulatory standards for the storage, transport and valorisation of other organic fertilizers, including products derived from processed animal by-products (BPAO) and fertilizers containing them.<br><br>All livestock activities referred to in the article 1st of the Regime Governing the Exercise of Livestock (RGEL), approved by the Decree-Law no. 214/2008, of November 10, as well as agricultural holdings using livestock effluents or the products derived therefrom in agricultural recovery, shall promote the application of the guidelines provided for in the Code of Good Agricultural Practices (CGAP). |
| **Traceability** | Decree-Law 134/2002, of May 14 | The present diploma establishes the system of traceability and control of consumer information requirements to which retail sales of fishery and aquaculture products are subject under Regulation (CE) no. 2065/2001, of the Commission, of October 22. |
| | Decree-Law 243/2003, of October 7 | Changes the Decree-Law no. 134/2002, of May 14, which lays down rules for the traceability and monitoring of consumer information requirements for the retail sale of fishery and aquaculture products. |
| **Labeling** | Decree-Law 26/2016, of June 9 | In addition to defining the general labelling requirements for non-pre-packaged foodstuffs, it also makes it compulsory for them to indicate the substances in their composition which may cause allergies or intolerances, implementation in the national legal system of the provisions of article 44th of the Regulation (EU) no. 1169/2011. |

| Area | Diploma | Description |
|---|---|---|
| **Labeling** | Regulation 1169/2011, of October 25 | On the provision of food information to consumers on foodstuffs, which changes the Regulations (CE) no. 1924/2006 and (CE) no. 1925/2006 of the European Parliament and of the Council and repeals the Directives 87/250/CEE of the Commission, 90/496/CEE of the Council, 1999/10/CE of the Commission, 2000/13/CE of the European Parliament and of the Council, 2002/67/CE and 2008/5/CE of the Commission and the Regulation (CE) no. 608/2004 of the Commission. |
| **Nutrition labeling** | Decree-Law 167/2004, of July 7 | The present Decree-Law transposes into national law the Directive no. 2003/120/CE, of the Commission, of December 5, relative on the nutritional labelling of foodstuffs. |
| | Decree-Law 54/2010, of May 28 | The present Decree-Law transposes into national law the Directive no. 2008/100/CE, of the Commission, of October 28, which changes the Directive no. 90/496/CEE, of the Council, relative to nutritional labelling of foodstuffs about recommended daily intakes, energy conversion factors and definitions. |
| | Regulation 1924/2006, of December 20 | Rectification to Regulation (CE) no. 1924/2006 of the European Parliament and of the Council, of December 20 of 2006, relative to nutritional and health claims on food. |
| | Regulation 107/2008, of January 15 | Changes the Regulation (CE) no. 1924/2006 relative to nutritional and health claims made on foods as regards the implementing powers conferred on the Commission. |
| | Regulation 109/2008, of January 15 | Changes the Regulation (CE) no. 1924/2006 relative to nutritional and health claims on food. |
| | Regulation 116/2010, of February 9 | Changes the Regulation (CE) no. 1924/2006 of the European Parliament and of the Council as regards the list of nutrition claims. |
| **Waste and heavy metals** | Regulation 2377/90, of June 26 | It lays down a community procedure for the establishment of maximum residue limits of veterinary medicinal products in foodstuffs of animal origin. |
| | Regulation 1881/2006, of December 19 | It sets maximum levels for certain contaminants in foodstuffs. |

### 3.1.2.1. Operation

The table 6 describes important legislation about operation in aquaculture with the respective diploma for consultation.

**Table 6 - Important legislation about operation in aquaculture.**

| Area | Diploma | Description |
|---|---|---|
| **Operation** | Regulation (CE) no. 853/2004, of the European Parliament and the Council, of April 29, with the latest version in April 1 of 2016 complementary to the Regulation (CE) 852/2004 with the latest version in April 20 of 2009. | Establishes rules for the production and marketing of live bivalve molluscs, echinoderms, tunicates and marine gastropods. |
| | Regulation 2074/2005, of December 5 | It lays down implementing measures for certain products under Regulation (CE) no. 853/2004, of the European Parliament and of the Council and for the organization of official controls under Regulations (CE) no. 854/2004, of the European Parliament and of the Council and no. 882/2004, of the European Parliament and of the Council, repealing the Regulation (CE) no. 852/2004, of the European Parliament and of the Council and changes the Regulations (CE) no. 853/2004 and (CE) no. 854/2004. |
| | Regulation 1019/2008, of October 17 | Changes the annex II of the Regulation (CE) no. 852/2004, of the European Parliament and of the Council, concerning the hygiene of foodstuffs. |
| | Regulation 558/2010, of June 24 | Changes the annex III of the Regulation (CE) no. 853/2004, of the European Parliament and of the Council, laying down specific hygiene rules for food of animal origin. |
| | Regulation 1276/2011, of December 8 | Changes the annex III of the Regulation (CE) no. 853/2004, of the European Parliament and of the Council as regards treatment to eliminate viable parasites in fishery products intended for human consumption. |
| | Decree-Law 11/2006, of January 19 | The present diploma changes the Decree-Law no. 79/2005, of April 15, adapting the Organic Law of the XVII Constitutional Government to the needs of coordination and monitoring of the transversal policy instruments and the objectives of administrative simplification. |

| Area | Diploma | Description |
|---|---|---|
| **Operation** | Decree-Law 74/2014, of May 13 | It establishes the rule of digital provision of public services, enshrines assisted digital attendance as its indispensable complement and defines the mode of concentration of public services in Citizen's Stores. |
| | Decree-Law 223/2008, of November 18 | Establishes the execution rules, in the national legal order, of the Regulations (CE) no. 852/2004 and 853/2004, of the European Parliament and the Council, of April 29, relative to hygiene of foodstuffs and hygiene of foodstuffs of animal origin, respectively. |
| | Ordinance 699/2008, of July 29 | The Regulation (CE) no. 852/2004, relative to the hygiene of foodstuffs, and the Regulation (CE) no. 853/2004, which establishes the specific rules of applicable hygiene to foodstuffs of animal origin, both of the European Parliament and the Council, of April 29, Do not apply to the direct supply by the producer of small quantities of primary products to the final consumer or to retail establishments supplying the final consumer directly or to the direct supply by the producer of small quantities of poultry meat and of lagomorphs slaughtered on the holding, to the final consumer or to retail establishments which directly supply the final consumer with this meat. |
| | Decree-Law 151/2005, of august 30 | Transposes to the internal legal order the directive no. 90/167/CEE, of the Council, of March 26, which lays down the legal framework for the manufacture, placing on the market and use of medicated feeding stuffs for animals, repealing the Ordinance no. 327/90, of April 28. |
| | Decree-Law 193/2007, of May 14 | The present Decree-Law transposes into the national legal order the directives no. 2005/8/CE, of the Commission, January 27, 2005/86/CE, of the Commission, December 5, 2005/87/CE, of the Commission, December 5, 2006/13/CE, of the Commission, February 3, and 2006/77/CE, of the Commission, September 29, that changes the directive no. 2002/32/CE, of the European Parliament and the Council, of May 7, On undesirable substances in animal feed. |
| | Decree-Law 67/2010, of June 14 | The present Decree-Law transposes into the national legal order the directives no. 2009/141/CE, of the Commission, of November 23, that changes the annex I of the directive no. 2002/32/CE, of the European Parliament and the Council, of May 7, as regards the maximum levels applicable to arsenic, theobromine, *Datura sp.*, *Ricinus communis* L., *Croton tiglium* L. e *Abrus precatorius* L. |

#### 3.1.2.2. Commercialization

The table 7 describes important legislation about commercialization in aquaculture with the respective diploma for consultation.

**Table 7 - Important legislation about commercialization in aquaculture.**

| Area | Diploma | Description |
|---|---|---|
| **Commercialization** | Order no. 6497/2014, of May 19 | So far, the VCN attribution process was carried out in the issuance of a social document - VCN attribution register, sent to the coordinating entities after the approval. The VCN assignment register contained information on the identification of the establishment and the operator, as well as on the authorized activities. This document was also updated, whenever there was any occurrence in the activity of the establishment (change of activities or change of operator responsible for the activity). |

### 3.2. Production Models

Based on the information gathered from the bibliography (books and scientific papers) and on the questionnaires performed to the aquaculture producers, it was defined the production models for the main species produced in Portugal on semi-intensive regime.

These models were designed for the automation of operations through production orders made for human intervention or equipment operations, combined with the different hydrological parameters to an intelligent production that can be adapted to the best cost-benefit.

### 3.2.1. Gilt-head bream

**Table 8 - Production model for gilt-head bream.**

<table>
<tr><td rowspan="1">Maternity (Filtration, UV)</td><td><u>Breeders</u><br>✓ Daily routines: measurement of hydrological parameters, feeding and cleaning of tanks.<br>✓ Induction of posture in breeding.<br>✓ Fertilization.<br>✓ Collection of eggs for incubation in the dark.<br>✓ Hatching.</td><td><u>Larval state</u><br>✓ Daily routines: measurement of hydrological parameters, feeding and cleaning of tanks.<br>✓ Transfer of newly hatched larvae to the larval culture room (larva day 0 to 4-5).<br>✓ First feeding with rotifers (larva day 3-5 until 19).<br>✓ Second feeding with artemia (larva day 17 to 54).<br>✓ Weaning (larva day 25) and third feeding with ration (larva: day 34).<br>✓ Swimblader test (Alevin day 60).<br>✓ Screening (Alevin at day 80 and with 0,5g).<br>✓ Vaccination (Alevin at day 90 and with 1g).</td></tr>
<tr><td>Pre-ongrowing</td><td colspan="2">✓ Transfer of juveniles (between 5 and 10g).<br>✓ Feed given by automatic feeders, react to the touch of the fish (feeding when necessary and avoids waste).<br>✓ Daily routine: measurement of hydrological parameters and check for dead (these are then frozen and collected by a waste company).<br>✓ Selection (with 1,5g and 5g).<br>✓ After each cycle, there is cleaning of the tanks.</td></tr>
<tr><td>Growing</td><td colspan="2">✓ Reception of the fish.<br>✓ Feed given by automatic feeders, react to the touch of the fish (feeding when necessary and avoids waste).<br>✓ Daily routine: measurement of hydrological parameters and check for dead (these are then frozen and collected by a waste company).<br><u>After reaching the weight between 400g and 650g:</u><br>✓ Starvation for 48 hours in winter and 24 hours the rest of the year.<br>✓ Fishing (by enclosure).<br>✓ Death by thermal shock.<br>✓ After each cycle, there is cleaning of the tanks.</td></tr>
<tr><td>Packaging and Sales</td><td colspan="2">✓ Selection by size and poorly formed.<br>✓ Washing with fresh water.<br>✓ Packing in polystyrene boxes.<br>✓ Storage in a refrigerator at 5°C.<br>✓ Delivery.</td></tr>
</table>

### 3.2.2. Parameters

*Breeding*

**Table 9 - Water parameters for the breeding phase for gilt-head.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 13 | 18 until 20 | 22 |
| **Salinity** | 27 | 33 until 35 | 37 |
| **pH** | 7.5 | 7.7 until 8.3 | 9 |

*Larval state*

**Table 10 - Water parameters for the larval state phase for gilt-head.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 13 | 20 until 24 | 26 |
| **Salinity** | 0.5 | 15 until 25 | 30 |
| **pH** | 7.5 | 7.7 until 8.5 | 9 |

*Pre-ongrowing*

**Table 11 - Water parameters for the pre-ongrowing phase for gilt-head.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 10 | 25 até 28 | 30 |
| **Dissolved oxygen** | 80%l | 90 % | Saturation |
| **Salinity** | 30 | 35 until 37 | 40 |
| **pH** | 7.5 | 7.7 until 8.3 | 8.5 |
| **$NH_3$ (mg/L)** | - | $0 < NH_3 < 1$ | 1 |
| **$NO_2^-$ (mg/L)** | 0 | 0 | 2 |

*Growing*

**Table 12 - Water parameters for the growing out phase for gilt-head.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 10 | 25 until 28 | 30 |
| **Dissolved oxygen** | 80%l | 90 % | Saturation |
| **Salinity** | 30 | 35 until 37 | 40 |
| **pH** | 7.5 | 7.7 until 8.3 | 8,5 |
| **$NH_3$ (mg/L)** | - | $0 < NH_3 < 1$ | 1 |
| **$NO_2^-$ (mg/L)** | - | 0 | 2 |

### 3.2.3. Seabass

**Table 13 - Production model for seabass.**

| | | |
|---|---|---|
| **Maternity (Filtration, UV)** | Breeders<br>✓ Daily routines: measurement of hydrological parameters, feeding and cleaning of tanks.<br>✓ Induction of posture in breeding.<br>✓ Fertilization.<br>✓ Collection of eggs for incubation in the dark.<br>✓ Hatching. | Larval state<br>✓ Daily routines: measurement of hydrological parameters, feeding and cleaning of tanks.<br>✓ Transfer of newly hatched larvae to the larval culture room (larva day 0-7).<br>✓ First feeding with artemia (larva between day 7 and 45).<br>✓ Weaning and second feeding with ration (larva: day 34).<br>✓ Disbonding (Alevin day 60).<br>✓ Screening (Alevin at day 80 and at 0,5g).<br>✓ Vaccination (Alevin at day 90 and with 1g). |

| | |
|---|---|
| **Pre-ongrowing** | ✓ Transfer of juveniles (between 5 and 10g).<br>✓ Feed given by automatic feeders, react to the touch of the fish (feeding when necessary and avoids waste).<br>✓ Daily routine: measurement of hydrological parameters and check for dead (these are then frozen and collected by a waste company).<br>✓ Selection (with 1,5g and 5g).<br>✓ After each cycle, there is cleaning of the tanks. |
| **Growing** | ✓ Reception of the fish.<br>✓ Feed given by automatic feeders, react to the touch of the fish (feeding when necessary and avoids waste).<br>✓ Daily routine: measurement of hydrological parameters and check for dead (these are then frozen and collected by a waste company).<br>After reaching the weight between 400g and 650g:<br>✓ Starvation for 48 hours in winter and 24 hours the rest of the year.<br>✓ Fishing (by enclosure).<br>✓ Death by thermal shock.<br>✓ After each cycle, there is cleaning of the tanks. |
| **Packaging and Sales** | ✓ Selection by size and poorly formed.<br>✓ Washing with fresh water.<br>✓ Packing in polystyrene boxes.<br>✓ Storage in a refrigerator at 5 ° C.<br>✓ Delivery. |

### 3.2.4. Parameters

*Breeding*

**Table 14 - Water parameters for the breeding phase for seabass.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 13 | 15 | 17 |
| **Salinity** | 25 | 27 | 38 |
| **pH** | 7.5 | 7.7 until 8.3 | 9 |

*Larval state*

**Table 15 - Water parameters for the larval state phase for seabass.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 13 | 20 until 24 | 26 |
| **Salinity** | 0.5 | 15 until 25 | 30 |
| **pH** | 7.5 | 7.7 until 8.5 | 9 |

*Pre-ongrowing*

**Table 16 - Water parameters for the pre-ongrowing out phase for seabass.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 10 | 25 until 28 | 30 |
| **Dissolved oxygen** | 80%l | 90 % | Saturation |
| **Salinity** | 30 | 35 until 37 | 40 |
| **pH** | 7.5 | 7.7 until 8.3 | 8.5 |
| **$NH_3$ (mg/L)** | - | $0 < NH_3 < 1$ | 1 |
| **$NO_2$ (mg/L)** | 0 | 0 | 2 |

*Growing*

**Table 17 - Water parameters for the growing out phase for seabass.**

| Parameters (water) | Minimum | Optimum | Maximum |
|---|---|---|---|
| **Temperature (°C)** | 10 | 25 until 28 | 30 |
| **Dissolved oxygen** | 80% | 90 % | Saturation |
| **Salinity** | 10 | 35 until 37 | 40 |
| **pH** | 7.5 | 7.7 until 8.3 | 8.5 |
| **$NH_3$ (mg/L)** | - | $0 < NH_3 < 1$ | 1 |
| **$NO_2$ (mg/L)** | - | 0 | 2 |

### 3.3. Validation of growth protocols

After the experimental activity, results were analysed.

#### 3.3.1. Sampling

With the measurement of the average weight every month we can see that the fish in each month trials grew in length and weight with both feeding methods.

**Table 18 - Initial and final weights and lengths of the three test months.**

| | Month | | |
|---|---|---|---|
| | **August** | **September** | **November** |
| **Initial weight (g)** | 162.56 ± 50.14 | 214.64 ± 55.72 | 294.89 ± 70.21 |
| **Final weight (g)** | 214.64 ± 55.72 | 294.89 ± 70.21 | 366.67 ± 86.08 |
| **Initial length (cm)** | 20.75 ± 3.97 | 23.25 ± 3.09 | 25.00 ± 3.29 |
| **Final length (cm)** | 23.25 ± 3.09 | 25.00 ± 3.29 | 27.00 ± 3.58 |

#### 3.3.2. Hydrological parameters

The parameters of Table I were measured and in August the mean of the pH was 7.7 and the salinity was 36.1.

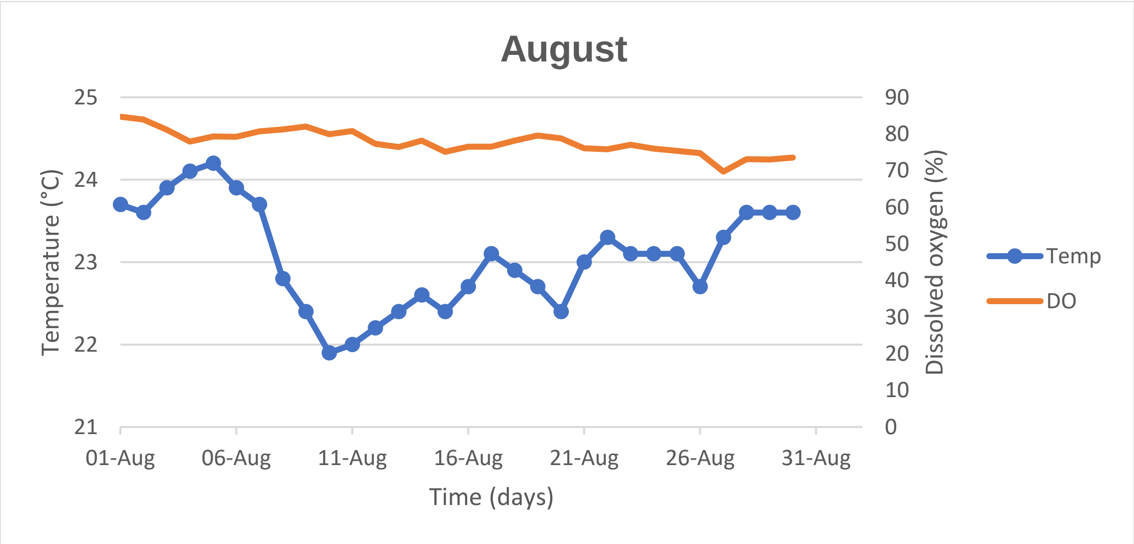


**Figure 16 - Temperature and dissolved oxygen ratio for August.**

In August the temperature varied from 21°C to 24°C, the dissolved oxygen was constant the all month varying from 70% to 80%.

In this graph (Figure 16) we can observe slightly that the temperature increases when the oxygen decreases.

In September the mean of the pH was 7.7 and the salinity was 33.9.

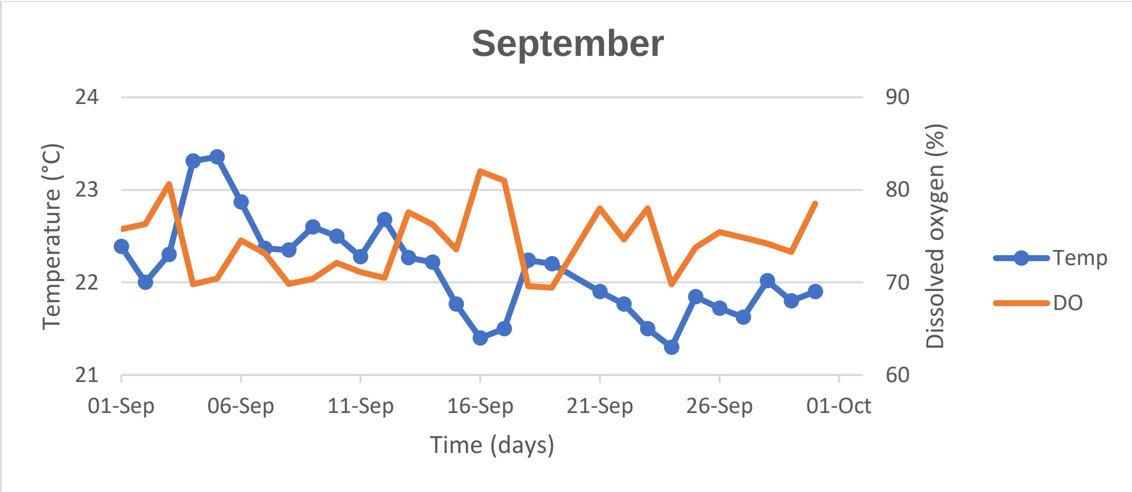


**Figure 17 - Temperature and dissolved oxygen ratio for September.**

During September the temperature varied from 21°C to 23°C, the dissolved oxygen varied from 70% to 82%.

In this graph (Figure 17) we can perfectly see that with high temperatures we have a decrease in dissolved oxygen, which suggests that fish with high temperatures have higher activity and higher oxygen consumption, this was very noticeable when the fish were being fed.

In October the mean for salinity was 35.6 and for pH level was 7.7.

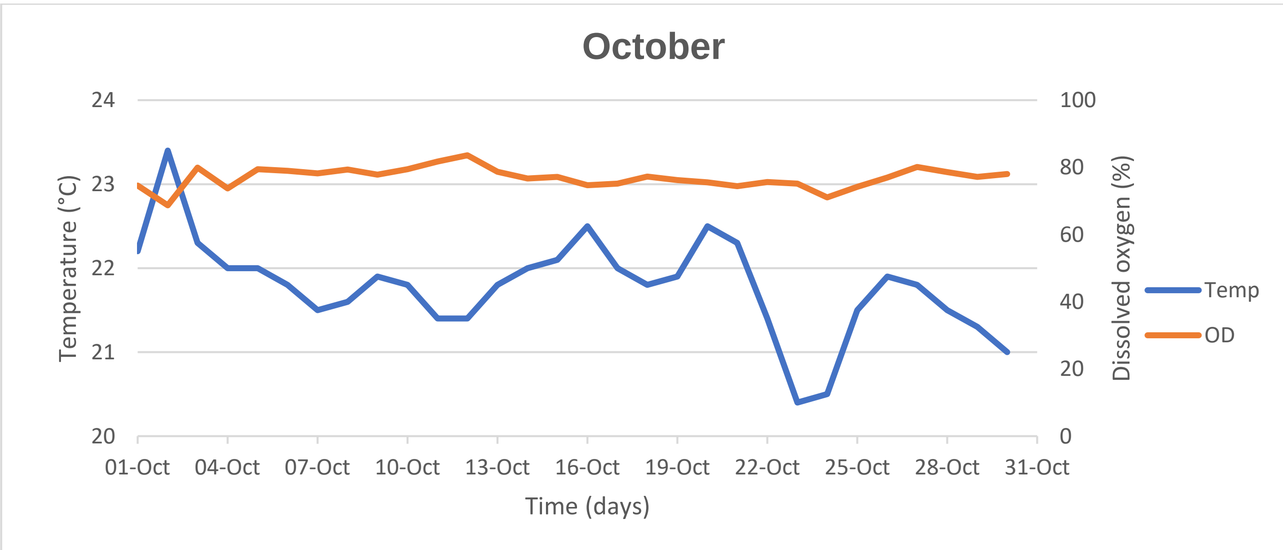


**Figure 18 - Temperature and dissolved oxygen ratio for October.**

During October the temperature varied from 20°C to 23°C, the dissolved oxygen varied from 70% to 83%.

In figure 18 we can see that there was a decrease in temperature in October, the dissolved oxygen remained constant and without great variations showing that the fish with low temperatures reduce their activity and consequently decrease the consumption of oxygen.

### 3.3.3. Growth rates

After the data collected and treated after the measurement of the biomass of the tank, we calculated the growth rates.

**Table 19 - Growth rates for the months August, September and October.**

| Month | Growth Rates | | | |
|---|---|---|---|---|
| | FCR | SGR | DGI | K |
| **August** | 1.04 | 0.93 %/day | 1.77 % | 1.72 % |
| **September** | 1.69 | 0.96 %/day | 2.03 % | 1.85 % |
| **October** | 1.26 | 0.68 %/day | 1.57 % | 1.79 % |

The best FCR was August with a value near to 1 which means that all the food given was converted to muscle, however it had the worst K meaning that they were growing less in weight and length.

The best SGR, DGI and K was always September. Despite of having the best specific and daily growth and the best fish condition (K), showing us that during that period increased in both length and weight, it had the worst food conversion ratio which means that the fish is converting food into fat.

In October it had the worst SGR and DGI that results from the low temperatures and according to the feeding table with less temperatures less amount of food given.

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## 4. Discussion

Legislation and licensing is still one of the obstacles to the successful development of the aquaculture sector.

Excessive and sometimes outdated legislation and lengthy licensing processes sometimes lead to high costs for parts of companies and is sometimes a major barrier to entry for new investors in the industry, so one of the objectives of Aquatropolis is to create a tool to help the producer regarding legislation and licensing with the construction of tables 3, 4, 5, 6 and 7 with the compilation of all the most important laws to take into account and with the presentation of all the regulatory entities that should be directed towards the approval of some title of use and exploitation of water resources.

To improve the sustainability of aquaculture with the capacity to maximize the semi-intensive production models, through advanced technological resources for the permanent and intelligent monitoring and control, in other words it is the automatic and real-time optimization of production models applied to a specie that is produced in a given location considering, therefore, the climatic and atmospheric conditions of the place of production.

Production models are address to specific species, in situations of unique and controlled production, throughout the project the knowledge about gilt-head and seabass production models was kept up to date (tables 8 and 13). It was also intended to systematize information on water quality parameters (Gilt-head - tables 9, 10, 11 and 12; Seabass – tables 14, 15, 16 and 17), characteristics of production systems and cultivation protocols.

Through the development of analytical and predictive models, fed in real time with data on water quality, Aquatropolis will make it possible to convert this information into systematic production models, to be interpreted and/or executed by the different profiles of managers, operators, technicians or specialists in aquaculture.

The productive efficiency of aquaculture industries is directly linked to the combined management of four main factors: food, energy consumption, ecosystem quality control and human resource management. The complexity of correlating the optimal points of these four factors in the production protocols can only be solved by transposing scientific knowledge to electronic platforms, designed to support producers in their day-to-day activities. The protocols of production are what allows us to ensure a systematic production cycle to reach the best product, faster, cheaper, with the least possible impact on the environment. Any and all disruption in the mentioned parameters will lead us to situations like: lack of appetite and consequent decrease of the rate of growth, diseases such as branchial lesions caused by the appearance of parasites and bacteria, reduction of reproduction rate, production of fry or larvae of inferior quality, among many others that will result in stress, malformations and mortality that will compromise the profitability of the industry and the guarantee of safety and food quality of the consumer.

Fish feeding is one of the most important factors in commercial fish farming because feeding regime may have consequences on both growth efficiency and feed wastage. Moreover, knowledge of the optimum feeding rate is important not only for promoting best growth and feed efficiency, but also for preventing water quality deterioration as a result of excess feeding. In intensive aquaculture systems, between 20 and 40% of the dietary dry matter is incorporated into the fish body and the remaining part is excreted. The proportion of uneaten/spilled feed ranges between 5 and 15%. The amount of faecal waste depends on factors such as feed composition, fish species and temperature (Amirkolaie, 2011). In this context, it is useful to know the optimum feeding rate of the cultured species and how feed efficiency, feed consumption and composition of flesh are affected by it (Eroldogna *et al.,* 2003). The accurate prediction of growth over a period is probably the most critical factor for accurately predicting the feed requirement in fish (Han *et al.,* 2011) and over the past few decades there have been many changes in feed technologies and feeding methods aimed at reducing the production of solid waste through uneaten/spilled feed (Amirkolaie, 2011).

From table 18 we can see that the fish grew more in September (in weight 80g and length 2 cm) when the food was given *ad libitum* (total food given – 4.5 kg) which studies with several fish species have revealed that with increasing feeding rate, the growth increases at higher ration levels and decreases at lower ration levels (Eroldogna *et al*, 2003). Since *ad lib* feeding of fish is difficult to carry out, one way to provide the daily energy requirement with minimal waste and high costs (price of September food – 4.46€) is to accurately estimate daily feed consumption using a bioenergetic model. An accurate bioenergetic model can be used to create precise feed charts that contribute to the economic and environmental sustainability of aquaculture. The bioenergetic model comprises three fundamental factors: feed level, fish size and water temperature (Han *et al.,* 2011).

With the feeding chart method (August and October) the fish grew less but not significant and it was the best method in respect of the quantity of food supplied (August – 2.2 kg and October – 2.9 kg) and price (August – 2.20 € and October – 2.84€) which goes against Cho,1992 and Olivetti de Mattos *et al*., 2016 that describes that most available feeding charts tend to over-estimate feed requirements resulting in overfeeding, poor feed efficiency under most husbandry conditions, feed waste, pollution of the environment and therefore economic disadvantages for fish farmers which constitute a major production cost in fish culture (Amirkolaie, 2011).

Several biotic and abiotic factors influence the growth of fish and water quality requirements of marine fish species farmed in Europe (turbot, *Scophthalmus maximus*; European sea bass, *Dicentrarchus labrax*; cod, *Gadus morhua*) have been examined for development of land-based rearing systems. In turbot, temperature and oxygen and their interactions exhibited long-term effects on growth but there are differences between fish from different geographic areas (Person-Le Ruyet *et al*., 2004). Temperature plays an important role in governing growth of sea bass and gilt-head via its effects on feeding and metabolism. As we can see in figures 16, 17 and 18 thermal effects interact with both external (oxygen and food supply) (Person-Le Ruyet *et al.,* 2004), when there were higher temperatures there was accelerated metabolism, increased oxygen consumption and increased feed intake and we could see that in September when food was given *ad libitum*. The peak of feeding preceded the acrophase of water temperature (the time when the temperature as a peak), which is in agreement with the phase relationship found by Sánchez-Vázquez *et al*., 1998, but differs from the findings of Anthouard *et al*., 1993, who noted a direct relationship between daily fluctuations in water temperature and feeding peaks (Azzaydi *et al.,* 1998). The DO is one of the most important abiotic factors determining the growth and survival of fish in both the wild and aquaculture, it increases feed consumption, feeding efficiency, metabolism and growth in fish. Numerous studies have shown increases in oxygen consumption after feeding due to the metabolic cost of digesting and assimilating the nutrients from the feed (Zhang *et al.,* 2011). According to figures 15 and 16 the levels of dissolved oxygen where below the minimum for the species that could have a significant impact on feed intake and growth but according to Bermudes *et al.,* 2010 there have been diverging results on whether suboptimal dissolved oxygen levels have a negative effect on feed efficiency or not, which can explain why we didn't have any negatives effects in growth and feed intake.

Fish feeding schedules are frequently designed by hatchery operators, which might give preferential consideration to staff working hours rather than biological or technological factors. Feeding of fish continues to be seen as an "art form" with the farmer, not the fish, deciding when and how often they are fed and estimating "satiety or near-satiety" (Olivetti de Mattos *et al*., 2016).

Both feeding methods as advantages and disadvantages. Feeding charts has as advantages the low feed cost, little food waste which at the end we can reduce one of the main costs of an aquaculture company and the correct prediction of the final weight of the fish that we have in the tank; this method has as disadvantage: the fish is not completely satisfied. The *ad libitum* method has as advantages a better and bigger growth of the fish and a better satiation of the fish; it has as disadvantages the waste of food, consequently more polluted water and poorer water quality, it has higher costs at the end and we don't know how much weight of fish we do have unless we measure them.

Feed conversion ratio is one of the most important principles for a fish farmer to understand. The cost of fish feed claims over half of the total budget for most fish farms. Consequently, it is very important for a fish farmer to get the optimum performing feed for the most economical price. The best value for FCR is 1 which means when giving 1 kg of feed the fish is converting it all to muscle, high FCR values means that the fish is overweight.

Feed Conversion Ratio (FCR) of European sea bass under commercial conditions in the Mediterranean region is often poor and highly variable. From a practical point of view, sea bass farmers have fed their fish according to feeding charts issued by the feed suppliers. These charts prepared under idealized environmental conditions are not always appropriate as stocking rate and size distribution of fish differ from one farm to another. In addition, these charts, which are adapted for water temperature or size of fish, may not be established accurately (Eroldogna *et al.*, 2003), which goes against the results of this study where the best FCR was when using the feeding chart as a method (August and October).

Specific Growth Ratio (SGR) and Daily Growth Index (DGI) are similar, they both predict the weight gain during a period of time. The lowest value was in October meaning the fish grew less due to low temperatures so the demand for food was less. Although widely accepted as the standard method, SGR is the mathematically most unsuitable function to describe fish growth when using both long and short-term data. Due to its exponential background, it must underestimate all intermediate data points. Its exponential form also grossly overestimates predicted body weight greater than the final body weight. For sure, the assumption of continually exponential growth in fish can be stated incorrect (Dumas *et al.,* 2010). Aquaculturists should consider using the absolute growth rate ($AGR = \frac{Final\ weight - Initial\ weight}{Time}$) or thermal growth coefficient ($TGC = \frac{Final\ weight^{\frac{1}{3}} - Initial\ weight^{\frac{1}{3}}}{Temperature \times Days}$), which are both easy to apply and achieve better prediction results and better fit to intermediate data and results are equally simple to compare and to understand. The AGR is a quick and easily applicable way to classify growth. It is widely accepted for comparing results in nutrition and growth studies. The disadvantage of both AGR and SGR is that comparison is only possible if fish are exactly of the same age, because the function peculate the natural rhythm of growth of fish during different life stages, which is not the case in the TGC (Lugert *et al.,* 2014).

Fulton's condition factor (K, the ratio of total weight and $L^3$) then characterizes the abundance of energy reserves relative to structure. Keeping the condition index (K) constant while growing in length requires increasing energy reserves proportionally to the structure (Bavčević *et al.*, 2010). The structure can only increase, while K can fluctuate. The low K were in August and October because when a fish requires more energy than it receives from feed it uses energy from its reserves for sustenance, thereby reducing K and, consequently the total weight of the fish. If a fish receives more energy than needed for its metabolism, it stores the energy into energy reserves, thereby increasing K to a maximum for its length. Maximizing K for a given size by definition also maximizes the total weight of the fish for that size which was what happen in September. We define the weight increase potential of a fish of given length and condition (K) as the maximum attainable increase in weight in a given period. The potential can be fully realized only when length increases by a maximum possible amount, and K becomes maximal. It follows that any retardation in growth that cannot be compensated for in a given period leads to a loss of total weight increase potential for that period. In aquaculture settings, loss of weight increase potential implies that fish will have to be cultured longer to attain the desired weight (Bavčević *et al*., 2010).

The fish farms require a healthy environment that implies a continuously monitor and check of its environmental parameters. In a semi intensive aquaculture traditional fish farms are till now mostly manually operated and their management requires the farmers to pay attention to a lot of factors for success, without having full control over the environment (Cario *et al*., 2017). The traditional YSI Professional Series probe has as advantages the lower cost and it permits to measure the hydrological parameters in different parts of the tank because of its extreme flexibility (YSI, 2017); as disadvantages the large amount of time it takes to measure the parameters in all tanks of an aquaculture. The Hydrolab DS5 Water Quality Multiprobe has as advantages to be constantly in the tank so save time in measure the parameters in all tanks and if there is any change in any parameter the fish farmer can take a faster action or how it has a flexible design it can be use in either profiling or long-term deployments, constantly monitorization and measures up to 16 parameters simultaneously (some sensors provide multiple parameters), seven universal sensor ports that allow custom configuration, it can be connected to an alarm system warning the fish farmer of some change saving the company of some damage (OTT, 2017); has as disadvantages the higher costs not only in the probe but all the equipment and software designed for its proper functioning and it is fixed in one only location of the tank so it is hard to see the fluctuations during the day in the all tank.

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## 5. Conclusion

After completing this study, we can conclude that relatively to the legislative question in Portugal in the aquaculture sector there are still many gaps to fill, making legislation easy to reach and understanding by the competent entities will be an additional assistance tool for the producer.

The production models with all the scientific knowledge transposed to technological platforms will be an added value for the producer in the tasks of his daily life, thus filling some gaps and completing the emerging needs of aquacultures.

We realized that to reduce feed costs the best feeding method to use is the feeding table with the help of the Hydrolab DS5 Water Quality Multiprobe with a constant measurement of all parameters of the water providing a faster action in case of any change of water quality in their aquaculture.

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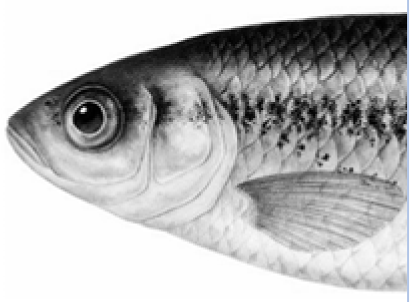
## 7. *Annex/Appendix*

**Annex I –** Fish farms' questionnaire.



**Grupo I – Caracterização da produção**

1. Caracterize e enumere os recursos humanos e perfis necessários para garantir a operação da aquacultura?
   ***Objetivos:*** *identificar quantas pessoas trabalham na empresa, quais os perfis, quais as lacunas, …*

2. Descreva-nos o ciclo de produção por espécie
   ***Objetivos:*** *identificar as fases do processo produtivo, identificar* inputs e outputs de cada uma das fases e duração das mesmas.

3. Descreva-nos o layout da sua produção.
   ***Objetivos:*** *compreender o número, tipo de tanques, características técnicas de equipamentos utilizados na produção, compreender qual a capacidade da produção e qual a capacidade que está em uso.*

4. Quais são os riscos e as incertezas associado a cada uma das fases?

5. Como descreve o processo de abastecimento de água para a produção
   ***Objetivos:*** *compreender a origem, quais os recursos técnicos para a sua captação, como é que a água é tratada, quais as características dos equipamentos utilizados?*

**Grupo II – Detalhe do processo produtivo**

6. Para cada uma das fases de produção, identifique quais são:
   - **a.** Atividades / tarefas

   - **b.** Protocolos de higienização

c. Protocolos de manutenção

d. Protocolos de alimentação

e. Protocolos de vacinação

f. Protocolos de manipulação (seleção- peixes tortos, triagem- dividir por tamanho)

7. Fazem seleção? Se sim, quais são os processos levados a cabo e destino para os exemplares não conformes?

8. Em termos de controlo de produção, quais são os processos e tecnologias utilizadas para a gestão dos lotes? Como gerem os protocolos e os modelos de produção?

9. Fazem triagens? Continua a ser o mesmo lote? Continua a ter a mesma designação?

10. Para cada uma das fases do ciclo produtivo, quais são os parâmetros que são monitorizados e com que frequência:
    a. Físico-químicos

    b. Parâmetros biológicos

11. Para cada uma das fases de produção, identifique quais são as condições ótimas e quais as condições que conseguem obter:

    a. Amónia e nitritos

    b. Temperatura

    c. Oxigénio dissolvido

    d. Salinidade

    e. Outras condições que se considerem relevantes (*blooms de microalgas*)

12. De que modo é que medem ou monitorizam o desenvolvimento da biomassa?

13. Que outros parâmetros gostavam de monitorizar / ou ter controlo para incrementar a garantia de qualidade, segurança e eficiência produtiva?

**14.** Quais são os recursos tecnológicos utilizados para garantir a monitorização e o registo e a manutenção dos dados em cada uma das fases do processo?

**15.** Quais são os indicadores (*Key Performance Indicators - KPI's*) que são utilizados para a gestão de cada uma das fases de produção?

**Grupo III – Caracterização dos processos administrativos**

**16.** Para cada uma das fases, identifique quais são os registos legais obrigatórios e quais os registos que têm de ser mantidos?

**17.** Estão sujeitos a auditorias?

**18.** Quais são os processos, registos e ferramentas utilizadas para dar resposta aos requisitos normativos de rastreabilidade?

**Grupo IV – Posicionamento de Mercado**

**19.** Quais são os critérios para a tomada de decisão para a Pesca?

**20.** Qual é o destino do pescado produzido? Quais são os canais de distribuição e comercialização?

**21.** De que modo é que conduzem a produção para que o pescado chegue ao mercado no momento em que pode ser mais valorizado?

**22.** Como descreve o seu posicionamento relativamente à concorrência em termos de processos e recursos tecnológicos? Em que ponto é que gostaria de estar?

**23.** Quais são as principais necessidades para que a vossa produção seja mais competitiva?

**24.** Identifique quais são os seus concorrentes.

**25.** Quais são os objetivos a curto-medio prazo para a vossa organização?