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Epidemiological status of Mediterranean farmed fish

Diseases in European seabass and gilthead seabream in European and Mediterranean farming: a comprehensive approach for a more efficient policy making

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1. INTRODUCTION

As many other farming activities, aquatic animals and particularly finfish can be affected by several infectious and non-infectious diseases. The specific diseases and pathogens or other causes triggering the development of these diseases, since there are usually multifactorial, can be related to a number of different factors: characteristics of the infectious agents, affected fish species or genetic susceptibility, environment and environmental conditions, geographical parameters, and farming systems amongst others.

2. DISEASES OF EUROPEAN SEABASS AND GILTHEAD SEABREAM

2.1 Available information

Diseases affecting European seabass and gilthead seabream have been widely reported in different technical and scientific papers, books and also in different professional / technical and scientific meetings.

The amount of published scientific and technical papers is not as high as in other farmed finfish species such as salmon or rainbow trout but the core of available information is really relevant ([Table 1](#)).

Table 1: number of results recovered from the search tools Google Scholar and Web of Science

Searching Google Scholar (April/2019)

Keywords: disease, *Sparus aurata*: 23.500 results

Keywords: disease, *Dicentrarchus labrax*: 22.700 results

Keywords: disease, *Salmo salar*: 58.000 results

Keywords: disease, *Oncorhynchus mykiss*: 65.400 results

Searching Web of Science (April/2019)

Keywords: disease, *Sparus aurata*: 2.048 results

Keywords: disease, *Dicentrarchus labrax*: 1.960 results

Keywords: disease, *Salmo salar*: 6.487 results

Keywords: disease, *Oncorhynchus mykiss*: 9.853 results

2.1.1 Scientific papers and books

Many of the papers on Mediterranean fish diseases focuses on single case description or on a specific pathogen or on a specific aspect of the disease (pathology, pathogen characterisation, diagnostics, treatment, immune response), and only few of them provide a summary of all the diseases with an integrative epidemiological approach. One of the first integrative review paper on diseases in Mediterranean farmed marine fish species with specific references to European seabass and gilthead seabream was published in 1998 by Rodgers and Furones, although a first pioneer overview of the diseases affecting gilthead seabream with a particular focus on Eilat,

Israel, had been already published by Dr Ilan Paperna and coworkers in 1977. A list and descriptions of diseases of European seabass and gilthead seabream was also included in the FAO Manual on Hatchery Production of European seabass and Gilthead seabream - Volume 1 (Moretti et al, 1999). Some diseases of gilthead seabream and European seabass were also presented in the marine version of the booklet “What should I do” (EAFP, 1997). Some years later, an integrative approach was also adopted in two chapters on diseases and health in sparidae and mainly in gilthead seabream (Colorni and Padrós, 2011) and in European seabass (Sitjà et al., 2014) and most recently in a book regarding health aspects in both species (Patarnello and Vendramin, 2017). All three works nicely summarize and detail the current knowledge of the diseases of these species. The fact that many diseases and the pathogens affecting these species are also addressed in other general books on fish diseases it should also be stressed. The updated list of diseases and pathogens affecting European seabass and gilthead seabream is available in chapters and books previously mentioned, so duplication of information that would increase the length of this document unnecessarily will be avoided.

It is also important to highlight that, as in other terrestrial and aquatic animal diseases, emerging diseases or new disease conditions may appear. These emerging disease conditions are normally communicated in professional meetings and conferences and require some time to be fully characterised and published in regular paper or included in the review papers. This was, for example, the case of winter disease syndrome, petequial rash and *Piscirickettsia* infections (Padrós et al 1996; Padrós and Zarza 2007, ; Zrnčić et al., 2015) and more recently, sudden death syndrome in European seabass and mortalities associated to strong gill pathology in European seabass and gilthead seabream (Padrós, personal observations, Bosch-Belmar et al., 2017), the new pathogens associated with epitheliocystis (Qi et al, 2016; Seth-Smith et al, 2017) and the emergence of *Enterospora* (Palenzuela et al, 2014) and enteric disorders (Padrós, personal observations).

2.1.2 Meetings, courses and projects

European seabass and gilthead seabream diseases started to be specifically addressed in meetings and courses in the '90s. The first ‘Fish Health Management in European seabass and Gilthead seabream Farming’ seminar on Disease Diagnosis and Control of Mediterranean Cultured Fishes was held in Malta in 1996, organized by Ecomarine Ltd. (Greece), and the Laboratory of Aquaculture and Artemia Reference Centre (Belgium), in association with European Commission (Leonardo Program), the Scottish Aquaculture Training Association, and AQUA TT U.E.T.P. Limited. This first initiative was organised when Mediterranean Aquaculture was still growing (total gilthead seabream and European seabass production around 50,000 tn (source: FEAP), only a 12.5% compared with the estimated production of 399,724 tn in 2017 (sources: FEAP, FAO, Apromar). Two years later, a short practical course on Fish Health Management was held in Udine (1987). In 1998, another short Practical Course in Fish Health Management in Mediterranean aquaculture was organized by the National Centre for Marine Research in Athens (Greece) in the context of a Leonardo Da Vinci Program and finally in 1999, a MEDIS initiative on fish diseases and disease management of Mediterranean marine aquaculture was organized in Rhodos (Greece). These were the first meetings specifically addressing Mediterranean fish diseases. In 2003, an International Advanced Seminar: “The use of veterinary drugs and vaccines in Mediterranean Aquaculture” was organized by CIHEAM

(International Centre for Advanced Mediterranean Agronomic Studies) in Izmir, Turkey. Later, in 2004, an update on many different aspects of the diseases was also presented in the Fish Disease Diagnosis and control in Mediterranean Aquaculture advanced course organised by CIHEAM in Santiago de Compostela. In 2011, the Workshop “Health management - the prerequisite for efficient breeding of Mediterranean species” was organized by the Croatian Veterinary Institute on 11 September 2011 in Split, Croatia, and also, very recently (2018) a training course on “Diagnostics approach to parasites of European seabass and gilthead seabream” was organised by the University of Udine in collaboration with the University of Bologna in the context of the EU-funded project ParaFishControl.

Diseases of gilthead seabream and European seabass were also regularly addressed at the level of the European Union Reference Laboratory (EURL) for Fish and Crustacean diseases. Since 2013, different updates on fish disease situation in the Mediterranean basin were presented in the annual meetings and workshops, with Niccolò Vendramin as the contact person and coordinator of data collection by the means of questionnaire to specialists and presentation of the results.

It should also be remarked that other documents, courses and meetings were and are regularly organised at regional or local level. European seabass and gilthead seabream diseases are usually well represented at the SIPI annual meetings in Italy (<http://www.sipi-online.it/convegni/default.aspx>) and specific issues on Mediterranean finfish diseases were particularly addressed in several years (**Table 2**).

Table 2: SIPI meetings where particular issues on Mediterranean marine finfish species are addressed

<p>2003 (X Convegno Nazionale della Società Italiana di Patologia Ittica S.I.P.I., 9-11 October 2003, Teramo: Tavola Rotonda “Principali patologie nella maricoltura mediterranea – Main diseases in Mediterranean mariculture”),</p> <p>2008 (XV Convegno Nazionale della Società Italiana di Patologia Ittica S.I.P.I., 22-24 October 2008, Erice (TP): Workshop “Acquacultura mediterranea: aspetti normativi e sanitari a confronto – Mediterranean aquaculture: regulatory and health issues compared”),</p> <p>2015 (XXI Convegno Nazionale della Società Italiana di Patologia Ittica S.I.P.I., 8-9 October 2015, Chioggia (VE): Workshop “Mare Adriatico: vecchi problemi e nuove sfide per l’acquacultura marina – Adriatic Sea: old problems and new challenges for marine aquaculture”)</p> <p>2017 (XXIII Convegno Nazionale della Società Italiana di Patologia Ittica S.I.P.I., 5-6 October 2017, Lecce: Workshop “Problematiche tecniche e sanitarie delle avannotterie marine – Technical and health issues in marine hatchery”)</p>
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In 2007, the “VI curso de ictiopatología práctica: prevención, diagnóstico y control sanitario en piscicultura marina mediterránea” (VI Practical Ictiopathology course: prevention, diagnostics and health control in Mediterranean marine finfish aquaculture) was held in the UAB (Barcelona).

In addition to these activities, gilthead seabream and European seabass diseases were frequently subjects of presentation and discussion in specialized fish fora, mainly in the

European Association of Fish Pathologists (EAFP) conferences and in specialized sessions in the European Aquaculture Society (EAS) conferences and in specific local and international conferences on viral diseases (Virus of lower vertebrates symposia), parasites (ISFP: international symposium on Fish Parasites, SOIPa Italian Society of Parasitology, SOCEPA Spanish Society of Parasitology), or fish health-related conferences (Fish & Shellfish Immunology conferences).

A particularly interesting initiative at national level (Spain) was also launched in 2007 as the [GESAC Project](#). GESAC (Guía para la gestión sanitaria en acuicultura/Health management guidelines in aquaculture) was designed as a collaborative work platform connecting fish health specialists and stakeholders and the different administrations in order to get information about the aquaculture diseases in Spain, epidemiological data and provide scientific criteria about these disease to promote better fish health policies. In this platform, gilthead seabream and European seabass diseases were very relevant.

We also would like to mention and highlight the value of specific personal initiatives in the dissemination of the knowledge and expertise of recognized colleagues such as Dr Panos Varvarigos and Dr Alain Le Breton with their participation in many of the aforementioned events and also in dissemination and efforts (websites).

Thus, we can consider that nowadays there is a substantial and updated body of knowledge and available information on the diseases affecting both species as well as a considerable number of specialists working on it.

2.2 Identification and scoring the significance of the diseases

With a historical perspective, the spectrum of diseases affecting European seabass and gilthead seabream in the Mediterranean has remained quite stable, with some changes mainly related to the impact of some of these in diseases during this period and the emergence of some new diseases.

Bacterial diseases have been traditionally one of the most frequently recorded and described problems in Mediterranean aquaculture. Rodgers and Furones (1998) already highlighted Vibriosis associated to different *Vibrio* species and Photobacteriosis (formerly Pasteurelosis) as the main bacterial problem in these two fish species along with Streptococcosis, *Tenacibaculum* infections, Epitheliocystis and Mycobacteriosis as bacterial diseases with some impact. Interestingly, these authors also highlighted the importance of nodavirus for European seabass production, presenting different aspects described in other species and stressing viral nervous necrosis (VNN) / viral encephalo-retinopathy (VER) as a real threat to Mediterranean aquaculture, at a time that the outbreaks associated to this virus were still relatively new in the Mediterranean production. The list of parasites appearing in the same review to be associated with European seabass and gilthead seabream was very similar to the list of parasites described in more recent reviews (Colorni and Padrós, 2011, Sitjà-Bobadilla et al., 2014); most of the parasite species that are considered significant for the gilthead seabream and European seabass sector were described in this review (*Sparicotyle* was still referred as *Microcotyle* and *Enteromyxum leei* as *Myxidium leei*), but some species such as *Enterospora* and *Lerneanthropus* were not cited yet.

Few years later, in the CIHEAM course in 2004, *Sparicotyle chrysophrii* was cited and highlighted as an emerging problem by Dr Alvarez Pellitero from IATS (CSIC) in his lectures on parasitic diseases. Dr Alvarez Pellitero also confirmed the increasing impact and relevance of *E. leei* infections in gilthead seabream. In the same course, Dr Bovo (IZSV), provided a complete update of the current available information on betanodavirus infection in the Mediterranean, indicating that VNN/ VER outbreaks were detected for the first time in the summer 1995 in many Mediterranean European seabass on-growing farms, with increasing mortality in the 1995-1998 period.

Importantly, in the Spanish GESAC project (2007-2009) previously described, the scoring system for Mediterranean fish diseases established in the European project PANDA (Permanent network to strengthen expertise on infectious diseases of aquaculture species and scientific advice to EU policy) was used. The results of this scoring process indicated that no disease was assigned in group 1 (high risk diseases /marine) but in group 2 (regional risk diseases, marine) the following diseases in gilthead seabream and European seabass were described: *Enteromyxum leei*, VNN & *Streptococcus iniae*. In group 3 (low risk diseases, marine) the pathogens assigned were *Sparicotyle*, *Tenacibaculum*, *Photobacterium damsela* subsp. *piscicida*, *Sphaerospora testicularis*. These results were similar, in general terms, to the results obtained in the following decade in similar exercises.

In the decade 2000-2010, although *Lernanthropus* and *Ceratothoa* were already described as crustacean parasites in European European seabass and gilthead gilthead seabream (Vagianou et al, 2006) and some reports on specific farms were recorded, these crustacean parasites did not yet represent a major problem for the finfish Mediterranean industry.

In the current decade (2010-2019) it seems that the ranking of the most important diseases in gilthead seabream and European seabass production is comparable to the previous one. In the periodic reports prepared by Dr Vendramin for the EURL for fish and crustacean diseases (years 2013, 2014, 2015 and 2016), the results of the different surveys based on questionnaires from different experts about the relevance and impact of the different diseases, results were quite similar, with VNN, Vibriosis, Gill flukes (*Sparicotyle*) and *Enteromyxum* as the diseases with the highest scores. Other diseases such as infections by *Photobacterium damsela* subspecies *piscicida*, *Tenacibaculum*, *Enterospora* or Petechial rash were also cited, but with lower scores. It should be noticed that for the first time, *Enterospora* and Petechial rash appeared in the ranking. In fact, Petechial Rash cases increased in the period 2007-2014 (Padrós, personal observations; Padrós and Zarza, 2014), while *Enterospora nucleophila* first complete description is also quite recent (Palenzuela et al., 2014)

Similar results (**Table 3**) with slight differences in importance amongst different areas in the Mediterranean were also obtained in the workshop on fish health in Mediterranean Aquaculture (EAFP, Las Palmas, 2015) as well as in an internal survey organized by the fish producers associations during PerformFISH proposal development (**Table 4**). In this survey, copepods (mainly *Lernanthropus*) gained momentum as important diseases mainly for European seabass.

Table 3: Mediterranean finfish aquaculture: disease relevance in the different areas, according to Vendramin et al (2016)

Rank	Western Mediterranean Area	Central Mediterranean Area	Southern Mediterranean Area	Eastern Mediterranean Area
1	VER-VNN	VER-VNN	VER-VNN	VER-VNN
2	Gill flukes: <i>Sparicotyle</i>	<i>Vibrio anguillarum</i> <i>Vibrio harveyi</i>	Gill Flukes: <i>Sparicotyle</i>	Vibriosis: <i>Vibrio anguillarum</i>
3	<i>V.anguillarum</i> , <i>V.harveyi</i> , <i>Ph. damsela</i> subsp. <i>piscicida</i> and <i>Aeromonas</i>	Gill flukes: <i>Sparicotyle</i>	<i>Enteromyxum</i>	<i>Photobacterium damsela</i> subsp. <i>piscicida</i>
4	<i>Tenacibaculum</i>	<i>Photobacterium damsela</i> subsp. <i>piscicida</i>	<i>Photobacterium damsela</i> subsp. <i>piscicida</i>	Isopod infestation: <i>Ceratothoa</i>
5	<i>Enteromyxum</i>	<i>Tenacibaculum</i>	Mycobacteriosis	Gill flukes: <i>Sparicotyle</i>

Table 4. Ranking of diseases according to significance for Mediterranean aquaculture (5; highly significant. The ranking was performed by the national farmer associations participating in PerformFISH

Diseases	Overall significance (1-5)	
	European sea bass	Gilthead seabream
Nodavirus /VNN	5	-
Copepods (<i>Lernanthropus</i> , <i>Caligus</i>)	4	-
<i>Photobacterium damsela</i> subsp. <i>Piscicida</i>	3	3
<i>Tenacibaculum</i> (Myxobacteriosis)	3	2
Vibriosis (<i>V. anguillarum</i> and others)	2	1
<i>Aeromonas veroni</i> and other <i>Aeromonas</i>	2(3)	-
Isopods (<i>Ceratothoa</i>)	2	-
Monogeneans, gill flukes (<i>Sparicotyle</i> , <i>Diplectanum</i> , others)	1	5
Myxosporidian parasites (<i>Enteromyxum leei</i> , other Myxosporidian parasites)	1	2(5)
<i>Rickettsia</i>	1	-
<i>Streptococcus</i>	1	-
Winter syndrome	3	-
Mixed enteric problems	-	2
Epitheliocystis	-	1
Petequal rash /red spot disease	-	2(3)
Lymphocystis	-	1(2-5)
Microsporidia	-	1

Finally, it is important to highlight that in the last 2-3 years, three H2020 projects (ParaFishControl, MedAID & PerformFISH) are in parallel addressing gilthead seabream and European seabass health with relevant components. ParaFishControl is covering a large number of significant parasitic diseases for European finfish aquaculture and particularly focusing on most of the relevant parasitic diseases in Mediterranean fish species. PerformFISH and MedAID have a workpackage each dedicated to fish health. The concurrence of these three projects in a similar period of time should be considered as a unique opportunity to contribute with relevant elements on epidemiology and disease policies in European seabass and gilthead seabream.

2.3 Specific and relevant epidemiologic aspects of some of the most relevant diseases

Different epidemiological aspects of the main diseases of gilthead seabream and European seabass described in the previous section were selected after an accurate review of the available literature on each disease and discussed amongst a panel of specialists from the different institutions (UAB, UNIBO, ARC, UNIUD & ULPGC) participating in WP3. These selected diseases were also the same diseases selected for the evaluation of the Health Risk Assessment exercise designed in Task 3.1.

The most significant epidemiological aspects for each disease are summarized as follows:

2.3.1 *Betanodavirus Infection*

Geographical distribution in the Mediterranean

Betanodavirus or nervous necrosis virus (NNV) has been recorded and isolated from outbreaks and also from asymptomatic European seabass since the first years of the 90's and more recently (Toffan et al., 2017) from gilthead seabream. The disease has been increasing its relevance and nowadays is considered the main disease for European seabass. Therefore, VNNV can be considered endemic in the Mediterranean, affecting farms in both East and West coasts (Toffan et al., 2017). Although there is not a homogeneous distribution across the Mediterranean due to different factors (concentration of the production in certain areas, different risk temperature ranges), VNN can be considered as an ubiquitous viral disease that is present in many other areas in Europe, Asia, America and Oceania.

Genotypes and reassortants

Amongst the different VNNV genotypes, RGNNV is the most commonly detected virus in European seabass, although SJNNV was also sporadically detected in some areas and more recently, the occurrence of reassortant strains (RGNVV/SJNNV and SJNNV/RGNVV) has been reported in the Mediterranean Sea. The emergence of the betanodavirus reassortant RGNVV/SJNNV strain extended the host range of VNN to gilthead seabream. At this moment, it mainly affects larvae, postlarvae and juveniles, but changes in the appearance pattern (clinical cases and significant mortality in on-growing sites) should not be discarded.

Virus survival in the environment

Although the virus stability is quite relevant at low temperatures, it is considered that VNNV stability is reduced at temperatures higher than 25°C and probably at this range of temperatures

the presence of the virus in carrier/reservoir fish may take a relevant role. The virus can be destroyed by desiccation but showed high resistance to several physical-chemical treatments and its inactivation require the use of high-powered biocides and adequate disinfection protocols.

Virus permanence in reservoir fish

Farmed European seabass and in some cases gilthead seabream but also other Mediterranean farmed species such as shy drum (*Umbrina cirrosa*) can be clinically affected by VNN. Wild fish such as dusky grouper (*Epinephelus marginatus*) or more recently Mediterranean moray eel (*Muraena helena*) can also be clinically affected and natural outbreaks are described in the Mediterranean. The permanence of the virus in the survivor fish after the outbreak (although at lower levels) is common, indicating that the risk of transmission of the virus from survivor fishes should not be underestimated. In addition, in many other wild fish species of the Mediterranean (*Mugil cephalus*, *Liza aurata*, *Seriola dumerili*, *Mullus barbatus* and probably many others) the virus can be detected (Ciulli et al., 2007).

Virus permanence in vectors

Some marine invertebrates (clams, mussels, polychaetes...) tested positive to the presence of the virus. This is not an unexpected finding as most of these invertebrates feed from the environment and can be filter feeders and therefore uptake viruses from the environment. NNV is not inactivated in the invertebrate digestive gland and infectious virus can be released via faecal matter and filtered water into the surrounding environment (Volpe et al., 2017). This is the reason why they can also be considered as vectors of the virus.

Transmission

Betanodavirus horizontal and vertical transmission has been demonstrated in several fish species, including European seabass. Horizontal transmission is the most common way of disease transmission through cohabitation with infectious fish or adding virus in the water. The potential presence of betanodavirus in virus-contaminated food sources is also suggested as an alternative way of horizontal transmission. *Artemia* and rotifers has been suggested to be a potential source for the introduction of betanodavirus in the larval culture. In fact, rotifers and *Artemia* can be experimentally contaminated with betanodavirus and the virus can be recovered and isolated from both organisms (Skiris and Richards, 1998) indicating that they can be carriers of the virus. However, no demonstration of viral replication in the rotifers or in the *Artemia* naupli was demonstrated. In the case of *Artemia*, a similar way of contamination it was suggested for LCDV (Cano et al, 2009). In broodstock, sometimes fresh or frozen fish or molluscs are used as a part of the diet. Considering that betanodavirus is a widespread fish disease and the virus has been detected in a wide number of fish species, the risk of horizontal transmission through contaminated fresh or frozen fish or shellfish should not be underestimated. Horizontal transmission risk through commercial feeds is much lower, as extrusion feed processes employ high temperatures (90-120°C) for several minutes. Evidence also exists for NNV infection of gonads (ovary and testis) and transmission from broodstock to eggs and larvae has been demonstrated in European seabass. It seems clear that the virus can be present in the ovarian fluid but is not so clear if virus can be found within the eggs. The use of disinfectants such as ozone in fertilized eggs seem to reduce the VNNV infection risk to larvae in other species (Mori et al, 1998).

Age

In European seabass, the disease causes high mortality rates in larvae and postlarvae (even affecting 100% of the stock). Mortality rates can also be relevant in post-larvae and juveniles and can also be significant in on-growing, where chronic mortalities are also frequently observed. In gilthead seabream affected by betanodavirus reassortants RGNNV/SJNNV strain, mortalities mainly occur in larvae and postlarvae but in juvenile, mortalities are lower than in European seabass and practically negligible in on-growing.

This particular susceptibility to the virus in larval stages and young fish and also the vertical transmission of the disease is particularly relevant for hatcheries and nurseries. The introduction of the virus not only can cause dramatic losses in the stock of larvae, postlarvae and juveniles, but also can affect and put in jeopardy the biosecurity of the stocks of juveniles produced in the hatchery that will be transferred or sold to pre-on-growing facilities and also can severely affect the future of the hatchery if the broodstock get infected by the virus.

Water temperature

Water temperature is a factor associated to the virus stability but also with the expression of clinical signs and mortality. In addition to the host specificity, genotypes also present particular specifications according to temperature. Particularly RGNNV is mainly expressed at high temperatures (25-30°C), while SJNNV genotypes affect fish species that usually live at lower temperatures (20-25°C). High mortality outbreaks in gilthead seabream larvae by reassortant RGNNV/SJNNV strain generally occur at 19-20°C.

Vaccines and vaccination

The situation on betanodavirus infection in the Mediterranean changed in 2018, as two new commercial vaccines against betanodavirus (RGNNV genotype) in European seabass (Zoetis/Pharmaq ALPHA JECT micro[®] 1 Noda and Hipra ICHTIOVAC VNN) received marketing authorization for Italy-Greece-Spain-Croatia and Portugal-Spain-France-Italy-Croatia-Greece-Cyprus, respectively. In the past, only experimental vaccines or autogenous vaccines were exceptionally authorized in some countries for limited use (special national authorization). According to manufacturer's indications, these vaccines (ALPHAJECT micro 1[®] Noda, ICHTIOVAC VNN) induce relevant protection rates: RPS final = 96% in 6 weeks after immunization (22°C, average weight at vaccination 15-16 g) using the same strain and RPS final = 79-91% as crossing protection using other betanodavirus field RGNNV strains for ALPHAJECT micro 1[®] Noda and RPS >60% in 42 days after immunization (22°C, average weight 15 g). Similar RPS (>60%) is achieved with ICHTIOVAC VNN after immunisation. High levels of protection (RPS final = 81%-82% according manufacturer specifications after 6 and 12 months, respectively at 22°C) are also achieved with ALPHAJECT micro 1[®] Noda. These values of efficacy and mid-long protection are comparable and even higher with levels achieved with other commercial vaccines against European seabass bacterial diseases and therefore are suitable to be integrated in vaccination plans in production cycles for at least one year.

There is a high prospect that other vaccines (including development from other experimental vaccines in the context of several EU-funded research projects (TargetFish, PerformFISH, MedAID) reach increased TLR and are also selected for commercial development and receive marketing authorization in the next years.

In this new scenario, vaccines and adequate vaccination programs including betanodavirus have become the heart of the betanodavirus management and strategies and policies for the future in on-growing. New vaccines including different genotypes and reassortants, with higher protection levels and during longer periods and bigger fish at risk, together with multivalent vaccines covering viral and bacterial diseases and new vaccination programs will definitely overpass the hurdle that betanodavirus infections impose for the development of Mediterranean aquaculture.

2.3.2 *Vibrio anguillarum*

Geographical distribution in the Mediterranean

Vibriosis caused by members of the genus *Vibrio* can affect a large number of wild and farmed species mainly in marine environments but also in other aquatic environments in the world. *Vibrio anguillarum* infections have a worldwide distribution as well as a wide distribution across the Mediterranean, that makes it endemic in this area.

Amongst the different O serotypes described, O1 and O2 are the two main serotypes usually isolated. Virulence may also vary according to the strains. In addition, not all the *V. anguillarum* isolates are pathogenic to fish, as some environmental isolates are mostly non-pathogenic (Frans et al, 2011). Other relevant species of *Vibrio* (*V. alginolyticus*, *V. harveyi*, *V. ordali* and *V. vulnificus* (biotype2)) are frequently isolated in European European seabass and gilthead gilthead seabream.

Affected species and age

European seabass is the main species infected by *V. anguillarum* and it should be stressed that *V. anguillarum* does not significantly affect gilthead seabream. In fact, only very few cases of isolation of *V. anguillarum* from gilthead seabream or from other sparids are described in the literature or are reported from fish veterinarians. In contrast, European seabass is highly susceptible to *V. anguillarum* infection, from larvae to juveniles and on-growing fish, even in large size fish. Outbreaks in larvae and juveniles tend to be peracute or acute, with high mortality. In bigger fish, mortalities can also be severe but not so severe as in younger European seabass. Outbreaks associated with this bacterium have been detected since the early times of the development of the Mediterranean aquaculture. Although commercial vaccines are available, European seabass vibriosis is still considered an impactful problem in the Mediterranean aquaculture (see previous section on relevance of diseases).

Bacteria and environment

Like other *Vibrios*, *V. anguillarum* is a ubiquitous bacterium mainly in marine coastal and estuarine environments (Frans et al, 2011) but also in freshwater. In some cases, *V. anguillarum* can also be found as 'viable but not culturable' state. Therefore, hygiene, prophylaxis and immunoprophylaxis are crucial in the prevention and control of the diseases.

Transmission and pathogenesis

Infections mainly occur through horizontal transmission and invasion through the fish skin although oral route from other infected fish, contaminated water or food is also described. Chemotactic motility and cell adhesion capability (Frans et al. 2011) of *V. anguillarum* are associated with the early stages of pathogenesis of the disease at the epidermis and the

intestinal mucosa level. Once the bacterium has penetrated the fish barriers, other mechanisms such as protease and haemolysin activity, iron sequestering system and lipopolysaccharides play their role in the fast dispersion as septicemic infection and in the pathogenesis of the disease. Vibriosis by *V. anguillarum* is also clearly related to temperature. The bacterium grows fast particularly at temperatures 25-30°C. Outbreaks usually take place in spring and autumn, with some differences recording in different areas in the Mediterranean sea, when temperatures rise above 20°C or decrease to 20°C (although some outbreaks can also be detected in temperatures higher than 15°C). External factors, related to the development of stress and immunosuppression, are also related to the development of outbreaks.

Vaccines and vaccination

Luckily several specific commercial vaccines for Vibriosis in European seabass (Alpha Dip Vib, Alpha Dip 2000 and Alpha Ject 2000, Pharmaq/Zoetis, Ichthovac VR & Ichthovac VR/PD, Hipra, Aquavac Vibrio & Pasteurella, Intervet Hellas) are licensed in different European countries. All these vaccines present very high protection rates even after bath vaccination (RPS final >90%, 4 weeks after immunization) and also at mid-term (>90%, 13 weeks post vaccination, >70%, 17 weeks post vaccination with Ichthovac VR/PD). These high values in protection allow minimizing the risks of *V. anguillarum* infection and the losses associated with when these vaccines are applied following systematic vaccination programs. Other commercial vaccines for vibriosis by *V. anguillarum* are licensed in Europe, but only for use in rainbow trout.

Prophylaxis, control and treatment

Immunoprophylaxis is the main and recommended method to reduce the impact of *V. anguillarum* infections in pre-on-growing and on-growing systems, as the exposure to the pathogen in these environments and the probability for the stocks to become infected is very high. As it has been previously described, available commercial vaccines may play a significant role in the protection of the populations at risk. At hatchery and nursery level, vaccination efficiency in terms of protection is not so high as in bigger fish due to the limited immunocompetence of the young fry (< 1 g), the length of protection provided through bath vaccination, and the difficulties of employing IP vaccination in small fry. In this case, general hygienic measures under suitable health control practices to avoid contamination with pathogenic bacteria also apply for *V. anguillarum* control. Mitigating control measures such as the use of immunostimulants, prebiotics, probiotics or substances (natural or additives) to inhibit or reduce bacterial proliferation are also recommended to reduce and control the onset of the infection. Therapy with antibiotics using medicated feeds is the main therapeutic method used to control *V. anguillarum* outbreaks in European seabass in Mediterranean aquaculture. A complete and expanded review on this is available in PerformFISH Deliverable 3.3 “Best therapeutics practices for Mediterranean farmed fish”.

2.3.3 *Photobacterium damsela* subspecies *piscicida*

Geographical distribution in the Mediterranean

Photobacteriosis, otherwise known as pasteurellosis or pseudotuberculosis is caused by *Photobacterium damsela* subspecies *piscicida* (formerly *Pasteurella piscicida*). As *V. anguillarum*, this bacterium can affect a large number of wild and farmed species mainly in marine environment. Photobacteriosis has a worldwide distribution and it can also be

considered as an endemic disease for the Mediterranean finfish aquaculture.

This bacterium is characterised by a relative homogeneity in terms of biochemical characteristics and antigenic properties in the European strains. However, field results seem to indicate different responses in terms of virulence, and also in response to vaccination, suggesting that some differences between strains may exist.

Affected species and age

Gilthead seabream and European seabass can be affected by *Ph.damselae* subs *piscicida* at all ages, although the disease is more severe in larvae or juveniles, sometimes with acute mortalities >70% of the affected stock. In fish >50-100 g acute mortalities can also develop and as the fish weight increases, fish tend to be more resistant and chronic forms are more frequently detected.

Bacteria and environment

There is little information about the presence of this bacterium in the environment, but *Photobacterium* spp. are organisms that can be found in marine environments in the water, the sediment and also in many marine organisms with which the bacteria may establish symbiotic or pathogenic interactions (Labella et al, 2017). Interestingly, different species of *Photobacterium* spp. establish bioluminescent symbiosis in marine fish and other aquatic animals in the so-called 'light organs'. *Ph. d. piscicida* is also characterised by the particular resilience to survive phagocytosis by fish macrophages and also in fish cell lines (Acosta et al, 2009) and its ability to survive inside these cells. This particular capacity (survival inside marine organisms and even inside fish cells) should be taken into account on hygiene and prophylaxis practices for this disease. Like *V. anguillarum*, *Ph.d.piscicida* can also be found as 'viable but not culturable' state.

Transmission and pathogenesis

Photobacteriosis transmission mechanisms are still unknown but horizontal transmission (fish to fish and oral transmission) has been described; like with *V. anguillarum*, infections occur through invasion of the fish skin although oral route from other fish, contaminated water or food (including cannibalism, mainly for gilthead European seabass) are described. Vertical transmission has also been described for this pathogen (Romalde, 2002) through the seminal and ovarian fluids from carrier broodstock and this particular fact is crucial for prophylaxis and health control in the hatcheries. Adherence to cell surfaces (Acosta et al, 2009, Remuzgo-Martinez et al, 2014) has also been demonstrated. Photobacteriosis is a temperature-related disease. Temperatures above 18°C and particularly temperatures between 25 and 30°C are the normal thermal range for the development of Photobacteriosis outbreaks. This is the reason why Photobacteriosis outbreaks are normally detected in summer and early autumn in the Mediterranean. External factors, related to the development of stress and immunosuppression, are also related to the development of *Ph.d. piscicida* outbreaks.

Vaccines and vaccination

As for vibriosis, several commercial vaccines are available for Photobacteriosis in gilthead seabream (Ichtiovac PD, Hipra; Aquavac Photobac Prime, Intervet Hellas) and/or European seabass (Ichtiovac VR/PD, Hipra; AlphaDip 2000 and Alpha Ject 2000, Pharmaq/Zoetis and Aquavac Vibrio & Pasteurella and Aquavac Photobac Boost, Intervet Hellas) and licensed in different European countries. All these vaccines present lower and variable protection rates

than *V. anguillarum* vaccines (RPS final >60-90%, 4 weeks after immunization and >70%, 17 weeks after immunisation for Ichtiovac VR/PD). These protection values help to reduce the risks of *Ph. damselae* subsp. *piscicida* infection and the losses associated with when these vaccines are applied following systematic vaccination programs in the farms, but the risk of outbreaks with some mortality is not completely eliminated. Moreover, some field results indicate that the protection of these vaccines at mid-long term is progressively reduced, suggesting that new vaccine formulations with improved lasting protection are necessary. Particular cases of reduced or low vaccination efficacy in some stocks are also indicative of changing antigenicity. These two points are particularly addressed in PerformFISH WP3, Task 3 “Vaccines and vaccination”.

Prophylaxis, control and treatment

All the points previously described for *V. anguillarum* also apply to *Ph. d. piscicida*.

2.3.4 Sparicotyle chrysophrii

Geographical distribution in the Mediterranean

Sparicotyle chrysophrii is a common monogenean parasite in wild gilthead seabream (Reversat et al, 1992; Euzet & Noisy, 1997) and currently one of the most important parasitic problems in finfish farming across the Mediterranean (Vendramin et al, 2016). Problems related to the presence of this parasite were already recorded at the early stages of the development of the Mediterranean aquaculture (Paperna, 1987). Problems associated to this parasite (still referred as *Microcotyle chrysophrii*) start to be recorded from the Eastern to Western Mediterranean (Paperna, 1991; Faisal & Imam, 1990; Sanz, 1992). In the following years, problems in gilthead seabream farms associated with *Sparicotyle* increased with the increase of the production of Mediterranean marine finfish aquaculture.

Affected species and age

Sparicotyle chrysophrii is mainly found in gilthead seabream although it has been described in other sparids such as in sharpsnout seabream *Diplodus puntazzo* (Mladineo, 2006; Mladineo & Maršić-Lučić, 2007). Young fish are normally more sensitive to parasitic infection and can develop pathology at lower parasite intensity. A threshold value of 8 parasites / gill arch has been linked to high pathogenicity with gross lesions such as gill and systemic anemia (Mahmoud et al. 2014).

The parasite and the environment

Juveniles and adults live in the gills of the fish (Antonelli et al, 2010) and bundles of eggs are released by gravid adults into the environment. Each egg has two polar filaments (the distal filament with a hooked end) that tend to tangle between them and also tangle with other filamentous structures present of the aquatic environment or in submerged structures in the farm (gill filaments, nets, natural and artificial microfilamentous structures, algae, fouling, ...) and very frequently become attached to these structures. Oncomiracidia hatch after 5-14 days, usually in the darkness, and actively swim in the water column to find a new host within 52 h, although 54% of the oncomiracidia die after 12 hours and only 13% of them are alive after 24 hours (Repullés-Albelda et al, 2012; Sitjà-Bobadilla et al. 2006).

Parasites can be found in fish farms during all year round, although the developmental cycle is highly influenced by temperature, being shorter at higher temperatures, and making the lifespan of the parasites longer at lower water temperatures. Thus, the number of life cycles per period of time and consequently, number of new parasites produced is higher at higher water temperatures. Prevalence and abundance of parasites in the farms, however, does not exhibit clearly common patterns in all the Mediterranean areas and facilities. Although highest prevalence and high parasitic loads can be normally found at warmer temperatures, sometimes high numbers of infected gilthead seabream with higher intensity can be found in winter (Antonelli et al., 2010), with mortality outbreaks in farms with temperatures below 15°C (Sanz, 1992). In these conditions, gilthead seabream physiology and performance can be compromised due to sub-optimal temperatures. These differences between farms can be explained by particular differences related to their geographical location (water temperature regime, water currents) or farm management (total parasitic load, distribution and distances between cages, separation of different age batches, cleaning activities, antifouling strategies, frequency and efficiency of the treatments against *Sparicotyle* applied, ...). The impact on the farms and the fish can also differentiate at warm or low temperatures and with the presence of other diseases. This can be particularly critical when fish haematocrit is normally decreased, immune system is compromised (Henry et al, 2015) and fish are affected by conditions such as winter syndrome (Padrós, personal observations).

Transmission and pathogenesis

Farm internal reinfection represents the most frequent and relevant way of transmission in cage farms. In cages there is a constant reinfection and gilthead seabream is normally infected and re-infected from eggs and oncomiracidia released in the environment by other fish within the same cage or from the infected neighbouring cages. Fry/juveniles from nursery or pre-on-growing usually are not infected due to the specific rearing system, with complete physical separation of the production batches and the use of good water quality. Fry/juveniles become infected when they are introduced in cages in the farm in the vicinity of cages that are already infected. Wild fish or escapees may play a certain role as reservoirs of the parasites, but their relevance is reduced when the main parasitic load is already present in the rest of reared stocks in the farm. In tanks or ponds, although batches are physically separated, if complete tank or pond desiccation or appropriate hygienic sanitization and disinfection procedures are not applied between the different batches, reinfection is possible.

Prophylaxis, control and treatment

There are no vaccines available to prevent infection by *Sparicotyle*. In general, in cage farms the control of the parasitic level can be achieved through the implementation of the following strategies:

- Periodical cleaning/substitution of the nets and the rest of structures (ropes, cables, superstructures, mooring systems) to remove attached eggs and remove fouling to reduce the possibility of eggs attachment
- Periodical cleaning, in inland intensive farming systems, of the tanks and the submerged substrates/fouling able to entangle the eggs
- Periodical evaluation, substitution and/or antifouling of the nets
- Batch separation: new batches of juveniles should be introduced in cages separated from the cages already stocked potentially infected gilthead seabream, taking into

account also the local sea currents, to reduce the probability of reinfection within the farm

- Following measures (when possible)
- Biomass reduction and increase of water exchange, when feasible, to limit the oncomiracidial attachment to the fish host, based on its scarce resistance in the water environment
- Periodical assessment of the number of parasites per fish/per batch using a simple but efficient and validated scoring system
- Periodical treatments (bath) using licensed products and under close supervision. If possible, aggregated treatments (several neighbouring cages treated together in a short period of time) produce more efficient results as they prevent reinfections
- Evaluation of the efficacy of the treatments using the same validated scoring system
- Use of specific diets in specific moments to reduce the parasitic load in the farm. Most of the information about antiparasitics is already described in deliverable 3.3.

3. DATA AVAILABILITY ABOUT MEDITERRANEAN FISH DISEASES AND POLICIES

It is clear from the previous section that there is an acceptable level of knowledge of the gilthead seabream and European seabass diseases. This knowledge is probably not as extensive and thorough as in other species, mainly Atlantic salmon and other salmonids, but it is significant and forms a good basis for further improvement in different fields.

Different initiatives have been launched in the last years in order to score the severity of the different diseases. Although the methodologies used in these initiatives are not the same, the final conclusions of all of them are very similar, reinforcing the robustness of these results (see Tables 2 and 3 in section 2.1).

Epidemiology, epidemiological studies and health policies in general are highly dependent on the data collection. The complexity, deepening and trust level of the epidemiological studies is highly dependent on the availability and robustness of the data used in these studies.

3.1 Epidemiological Information: sources in human medicine

Epidemiological studies of diseases in humans are highly facilitated by extensive data-mining from highly reliable sources: from individual clinical surgeon practitioners, to health centres, clinics, hospitals, diagnostic laboratories and health surveillance networks. In addition to that, many human diseases (and mainly infectious diseases) are also under close inspection and follow up by public health institutions at national and also international level (WHO, UN, NGO's) and these institutions tend to work using similar or compatible data recording standards. In addition to that, specific surveillance networks for certain human diseases (mainly for OMS/government authorities noticeable diseases) and also data from specific research on human health are also widely available. A big part of these data is public or can be obtained from public databases with specific permissions, while most of them are obtained using strong quality standards, reinforcing the trustiness of these studies. Obviously there is also a considerable core of data obtained from studies run by private medical and pharmaceutical companies that

allowed new medical developments, but these data are normally confidential or with strict dissemination policies, to protect company know-how.

3.2 Epidemiological Information about diseases: diseases under official surveillance programs

Studies on epidemiology of animal diseases, including aquatic animals and finfish, usually do not have the same access to quantitative data as the studies in humans. Amongst the different diseases in the different species, diseases listed by the World Organization for Animal Health (OIE) or diseases under surveillance from government authorities take the advantage of the regular generation of data based on active and passive surveillance programs. At international level, the information on the epidemiology of these diseases is mainly managed by OIE and international organizations. In the EU, is the European Union Reference Laboratory (EURL) for Fish and Crustacean diseases, and also laboratories and officers from the government authorities of the different countries that collect this information. In some cases, certain data can be available from public databases and periodical reports. However, this information mainly applies to listed diseases or diseases under specific surveillance programs, but not for the non-listed diseases, for which the public available information is scarcer. In the latter cases, data availability is limited to specific research projects on specific diseases. Usually, data from these public-funded studies tend (but not always) to be available directly from technical documents (reports, dossiers) and scientific documents (papers in scientific journals), yet, most often information is available as aggregated data or statistical-processed results. General databases on these studies are not usually available and only rarely data are transferred to open document repositories and databases; however, repositories are not maintained for a long time.

In addition, the level of homogeneity and harmonization of the structure of these databases are much lower than for human diseases data. In contrast to human health, that is by nature, closer to the concept of 'public', animal health is much more based on private activities. Only for the most important diseases such as OIE listed diseases or the diseases listed in national and transnational regulations (such as EU regulations) there is a certain level of data availability. Concerning data from non-listed diseases, although some specific data from such diseases can be mined from scientific papers, most of this information is obtained and managed internally by the farmers, farming companies and external health experts and advisors. In these cases, the information and the data are not public as these are considered as confidential and also a part of the know-how of the companies.

In the particular context of the European seabass and gilthead seabream diseases it is very important to stress the fact that none of the diseases affecting both species are listed by OIE and also none of these diseases are considered in the EU regulations on aquatic animal health (Directive 2006/88/EC and Regulation (EU) 2016/429). Despite VNN was included in the past in the list of diseases of the OIE (last version, 5th edition of the Aquatic Animal Health Code, 2006) VNN is no longer included in the list of fish diseases. Therefore, no diseases of gilthead seabream and European seabass farming are nowadays under official specific surveillance programs.

To understand why these diseases are not listed, it is necessary to take into account the OIE criteria for listing aquatic animal diseases, according the Article 1.2.2.

3.2.1 The OIE regulatory perspective

According to the [Article 1.2.2.](#), OIE has set the following criteria for listing aquatic animal diseases:

Criterion 1) International spread of the pathogenic agent (via aquatic animals, aquatic animal products, vectors of fomites) is likely

As gilthead seabream and European seabass are currently farmed mainly in the Mediterranean area and no significant expansion in other areas is foreseen, the international spread can be considered as limited. This mainly applies to diseases such as *Sparicotyle* infection with a high degree of host-specificity. For the other diseases (VNN, Vibriosis, Photobacteriosis) it should be noted that the pathogens are already present (endemic) in many other areas in the world, and also affect many other species, including wild fish.

Criterion 2) At least one country may demonstrate country or zone free from the disease in susceptible aquatic animal

All the diseases of gilthead seabream and European European seabass can be considered as endemic diseases in the whole Mediterranean basin. Some of them also present holoendemic trends (affect mainly young animals) and others present hyperendemic trends (affecting all ages).

However, some geographical differences in the expression of the diseases can be identified for certain diseases. For example, the apparent lower incidence of nodavirus outbreaks on the East side of the Adriatic coast, or the lower incidence of *Ceratothoa* and *Lernenathropus* infection in the Western Mediterranean. All the important diseases and most of all other diseases and pathogens can be considered present across the Mediterranean. No countries or zones have actively or passively demonstrated to be free of the diseases so far and it is unlikely that this could happen in the future. Amongst the main reasons that hamper the implementation of the 'epidemiological zoning' concept in the Mediterranean are the following:

- a) The syncytial characteristics of the Mediterranean basin such as the fact that no geographical barriers between countries are present
- b) Coastal continuity: there are no oceanographic borders/limits between zones
- c) Mediterranean hydrodynamics
- d) High dissemination risks through wild fish populations as potential carriers/vectors for the pathogens
- e) Close contact between wild fish and farmed gilthead seabream and European seabass stocks, favoured for the use of sea cages as the main rearing system at the on-growing stages
- f) Migration of some wild species, including wild gilthead seabream and [European seabass](#)
- g) Cages and cage management implies a low bio-isolation level of the farmed stocks.

Under these circumstances, it can be assumed that is very difficult to keep free a zone or a country. The situation could be different if farming activity substantially changed towards recirculation systems, but this change is not probable in short or mid-term.

Criterion 3) No zoonotic diseases with severe consequences proved

Luckily, none of the diseases of European seabass and gilthead seabream can be classified as zoonotic diseases as there are not human diseases that can be directly associated to gilthead seabream or European seabass diseases. It is important to differentiate the fact that some marine aquatic organism such as some species of *Vibrio* (Austin, 2010) or environmental *Mycobacteria* has been sporadically associated with some problems as a consequence of wound infection contaminated by seawater or with gastrointestinal problems following the consumption of contaminated or spoiled fish (then, a problem of food hygiene, not fish diseases) in certain risk groups in humans. Although *Vibrio vulnificus* (biotype 2) has been occasionally isolated in European seabass (Fouz et al, 2010) no records of clinical cases in humans associated to consumption or contact with diseased or carrier European seabass have been described. Moreover, only certain serovars of *Vibrio vulnificus* biotype 2, specifically serovar E, have a relevant pathogenic potential. Amongst parasites, only *Anisakis* and some anisakids are considered as relevant zoonotic fish parasites in the marine environment where gilthead seabream and European seabass are reared. However, due to the particular transmission cycle of Anisakids and the fact that gilthead seabream and European seabass are fed on food pellets, the presence of *Anisakis* in gilthead seabream or European seabass is very unlikely. This fact was also verified in recent surveys (Apromar, 2012; Gustinelli et al. 2017) demonstrating that *Anisakis* is not a risk for farmed gilthead seabream or European seabass.

Criterion 4) The economic (and /or social) impact

The economic impact of any kind of disease on farmed animals (including gilthead seabream and European seabass) is obvious. The question is when to apply the concept “significant”. The preliminary assessment implemented in PerformFISH about the mortalities and losses associated to diseases clearly demonstrated that pathology is a significant issue in Mediterranean gilthead seabream and European seabass farming and pathology was considered one of the main hurdles to overcome towards increasing the production performance. However, although diseases are a critical handicap, at this moment none of them can be considered as really critical for the development of sector. In many cases, significance may also arise from the inability to control the disease by means of therapy, immunoprophylaxis or other preventive measures. Most of them are already available and it only requires improvement and much better implementation. The size of the sector at national or international level is also a very important issue to take into account to understand the interest of implementation of specific legislation or monitoring plans. Salmon industry is a strategic economic activity for countries such as Norway and Scotland and therefore, it is logical that a considerable number of resources and administrative efforts, including fish health and disease monitoring are invested in order to support this activity. In contrast, although European seabass and gilthead seabream aquaculture is a big activity in several Mediterranean countries, economic and social indicators such as volume of business, total production and number of employees are not as big as with livestock production.

To sum up, with the current information available, the profiles of the diseases of gilthead seabream and European seabass according to the OIE criteria are the following:

OIE Criteria	Yes/ No	Comment
1. International spread	Yes	Infectious diseases are easy to spread in a marine environment like the Mediterranean Sea

2. At least one country may demonstrate country or zone free from the disease in susceptible aquatic animal	No	Diseases occurrence profiles are very similar in all the Mediterranean
3. No zoonotic diseases with severe consequences proved	No	
4. The economic and /or social impact	Yes/No	The impact is evident, yet it appears to be assumable, controlled and reduced by the industry

As it can be noticed, at least 2 of the 4 criteria are clearly identified as “not fulfilled”.

3.2.2 The EU regulatory perspective

Similar approaches are also considered in the [EU regulation 2016/429](#) in chapter 2, article 5, section 3 concerning the diseases listed in Annex II. Diseases included in Annex II **should meet all the following five criteria**:

Criterion (i) Scientific evidence indicates that the disease is transmissible

Most of the diseases in gilthead seabream and European seabass are caused by viruses, bacteria or parasites and fulfil the criterion.

Criterion (ii) Animal species are either susceptible to the disease or vectors and reservoirs thereof exist in the EU

Again, gilthead seabream and European seabass are susceptible or vector/reservoirs for these infectious diseases.

Criterion (iii) The disease causes negative effects on animal health or poses a risk to public health due to its zoonotic character

Although virtually none of the gilthead seabream and European seabass diseases can be considered as zoonosis, however, negative effects of these diseases on the two fish species cannot be ignored. In any case, the term ‘negative’ does not identify the size of the impact.

Criterion (iv) Diagnostic tools are available for the disease

Although diagnostic accuracy can be improved, nowadays there are available tools for most of the infectious diseases in European seabass and gilthead seabream. However, there is no validation on techniques and procedures and consensus in their use for many of the diseases.

Criterion (v) Risk-mitigating measures and, where relevant, surveillance of the disease are effective and proportionate to the risks posed by the disease in the European Union

The risks posed by the different infectious diseases for gilthead seabream and European seabass farming industry are clearly limited, and can be kept under a reasonable and improved control by the industry. Moreover, risk-mitigating measures and surveillance is complex in the Mediterranean context as EU and non-EU countries share the same common space. In this scenario, it is really complicated to have risk-mitigating measures implemented at regulatory level.

All the different criteria are summarized in the following scheme:

EU Criteria	Yes/ No	Comment
(i) Scientific evidence indicates that the disease is transmissible	Yes	Nearly all infectious diseases are transmissible by nature
(ii) Animal species are either susceptible to the disease or vectors and reservoirs thereof exist in the EU	Yes	
(iii) The disease causes negative effects on animal health or poses a risk to public health due to its zoonotic character	Yes/No	Diseases by definition cause negative effects but no risks for public health are present
(iv) Diagnostic tools are available for the disease	Yes/No	For certain pathogens and diseases, diagnostic tools should be improved
(v) Risk-mitigating measures and, where relevant, surveillance of the disease are effective and proportionate to the risks posed by the disease in the European Union	Yes/No	

According this, there are doubts that three of the five criteria are fulfilled.

Diseases included in Annex II **should also meet at least one** of the following criteria:

(i) the disease causes or could cause significant negative effects on animal health in the EU, or poses or could pose a significant risk to public health due to its zoonotic character

Similar comments as for the previous criterion (iii)

(ii) the disease agent has developed resistance to treatments, which poses a significant danger to public and/or animal health in the EU

Although some resistance to antibiotics have been detected in some pathogenic bacterial strains of gilthead seabream and European seabass, no relevant multi-resistant bacterium, virus or parasite has been detected. In addition, Mediterranean farming industry is aware about this risk.

(iii) the disease causes or could cause a significant negative economic impact affecting agriculture or aquaculture production in the EU

Although it is unquestionable that some of the diseases of European seabass and gilthead seabream cause negative economic impact on the Mediterranean industry performance, none of these can be considered as limiting for the industry. Moreover, PerformFISH is addressing significant improvements in the prevention and control of the impact of diseases.

(iv) the disease has the potential to generate a crisis or the disease agent could be used for the purpose of bioterrorism

Definitely not

(v) the disease has or could have a significant negative impact on the environment, including biodiversity, in the EU

Although some of the diseases can also affect wild fish populations (nodavirus) no relevant negative impact in these populations has been detected.

All the different Annex II criteria are summarized in the following scheme:

EU Criteria	Yes/ No	Comment
(i) the disease causes or could cause significant negative effects on animal health in the EU, or poses or could pose a significant risk to public health due to its zoonotic character	Yes/no	Negative effects are present for most of the diseases but it is not clear if they can be labelled as 'significant'. No significant risk to public health.
(ii) the disease agent has developed resistance to treatments which poses a significant danger to public and/or animal health in the EU	Yes/no	Although resistance is detected in both bacterial and parasitic pathogens, resistance to treatments follows the general trends as in other animal production.
(iii) the disease causes or could cause a significant negative economic impact affecting agriculture or aquaculture production in the EU	Yes/no	Same comment as in (i)
(iv) the disease has the potential to generate a crisis or the disease agent could be used for the purpose of bioterrorism	No	
(v) the disease has or could have a significant negative impact on the environment, including biodiversity, of the EU.	Yes/no	Since now, no epidemiological records are described on pathogen significant negative effects on specific animal species or biodiversity in general.

In most of the different criteria, the word ‘significant’ is used. The interpretation of the meaning the word ‘significant’ in the sentences, as it was introduced in the beginning of this section, should be considered as a term to allow some flexibility for the legislators according to the specific circumstances of the impact of diseases in specific moments.

To sum up, according the EU regulations (based on OIE criteria), none of the Mediterranean diseases of gilthead seabream and European seabass fulfil all the criteria set. Although it is clear that they can be important in the scope of Mediterranean fish farming, as many other diseases in their corresponding areas (aquatic and terrestrial farming), at this moment there are not sufficient grounds to consider that the impact of these diseases should be rendered ‘significant’ in terms of specific EU regulations.

These aspects are particularly relevant and stressed in the points (26), (29), (30) of the preamble of the Regulation (EU) 2016/429:

(26) Not all transmissible animal diseases can or should be prevented and controlled through regulatory measures, for example if the disease is too widespread, if diagnostic tools are not available, or if the private sector can take measures to control the disease by itself. Regulatory measures to prevent and control transmissible animal diseases may have important economic consequences for the relevant sectors and may disrupt trade. It is therefore essential that such measures are applied only when they are proportionate and necessary, such as when a disease presents, or is suspected to present, a significant risk to animal or public health.

(29) Some transmissible animal diseases do not easily spread to other animals or to humans and thus do not cause economic or biodiversity damage on a wide scale. Therefore, they do not represent a serious threat to animal or public health in the Union and can thus, if desired, be addressed by means of national rules.

(30) For transmissible animal diseases that are not subject to measures laid down at Union level, but which are of some economic importance for the private sector at a local level, the latter should, with the assistance of the competent authorities of the Member States, take action to prevent or control such diseases, for instance through self-regulatory measures or the development of codes of practice.

All the previously described points support that the Mediterranean fish diseases, although important for the industry, should not be “listed” in the EU regulations such as other diseases of terrestrial and aquatic animal diseases.

3.3 Epidemiological information about diseases: diseases with no official surveillance programs

In this context, the existing public information on gilthead seabream and European seabass diseases with some basic information on the epidemiology is based on the references indicated in the previous section (books, review papers). In addition to this background based on scientific literature, it should be stressed that the vast majority of the epidemiological data on diseases is generated, recorded and managed as internal data in fish farms and/or companies and also by fish health practitioners/consultants and in some cases by diagnostic laboratories.

This epidemiological data in Mediterranean fish farming share the following characteristics:

1) Data from diseases, disease follow-up, mortalities and outbreaks are always considered as very sensitive information for the farms and companies. Companies tend to keep this

information and these data as confidential or at least, restricted. However, at the same time, this information is usually recorded and kept in databases in most of the finfish farms operating in the EU as all the European seabass and gilthead seabream farms in all the EU countries have similar surveillance and traceability systems as any other terrestrial livestock farms. Under these circumstances, the information concerning health aspects (mortalities, treatments, vaccinations) is available upon request to the responsible national or regional authorities, as for example, in the case of unexpected or unknown mortalities or significant changes in the epidemiology of specific diseases.

Disease occurrence in the farms is perceived as a sensitive issue, undesired or even 'taboo' for external diffusion. This is mainly due to their negative effects on the public and consumer perception if not reported adequately; previous negative experience with organizations against fish farming that they have used such information in a biased way to support their cause against the Atlantic salmon industry (<https://theferret.scot/pictures-diseases-farmed-fish/>) have proved the sensitivity of the issue. Biased or fake information can affect the prestige and reputation of the company and generate relevant negative commercial effects. Therefore, an engagement between transparency and confidentiality is always welcome.

2) Gilthead seabream and European seabass diseases and disease/health-related data collection and management may vary between companies and farms. Some farms use standard data management software or ad-hoc customized data management programs, sometimes adapted from salmon aquaculture. Usually farms of the same company use the same or similar management software to harmonize the information generated in each farm and facilitate the downstream analysis of this data. Therefore, there is not an agreed standardization of the type of information on health-related issues recorded amongst the different companies in the Mediterranean. The same problem has also been detected in MEDAID (Aguilera, personal communication) and the differences are even bigger amongst farms of different non-EU countries.

This was a relevant problem identified by the industry and there has been a strategic decisions to address the problem in PerformFISH; a specific interdisciplinary exercise for a better and more clear definition of the health-related Key Performance Indicators (KPIs) was implemented as a preliminary step for specific and harmonised benchmarking. PerformFISH is proposing a robust system for KPI definition, with important health –related KPIs and also operational welfare indicators along with a sophisticated new system for data collection and management based on a Virtual Research Environment (VRE). This system is developed in WP7 (KPIs Impact Assessment and Code of Conduct). The developments and results of this workpackage also will help with the harmonisation and standardisation of the health-related data collection procedures in the farms.

Confidentiality was particularly well-addressed in building the VRE in WP7. All KPIs including the health-related ones are introduced, stored, managed and analysed in an anonymous manner, using the same web cloud system (VRE) managed by the EU D4Science e-infrastructure in order to guarantee absolute confidentiality and protection of the KPI data. Thus, health-related data in PerformFISH are managed with a benchmarking internal view rather to an external pure epidemiological approach.

3) Data recording and transfer into the management software and supervision of these data requires an extra effort on human resources and consequently these resources invested have

also an impact in the economic balance of the farm/ company. It is obvious that for this reason, companies tend to consider the generated information as a part of their own 'know-how' and are often reluctant to share or distribute this information without an appropriate return/merit or revenue. The only way to facilitate the transfer of these data is in *quid pro quo* / tit for tat / win-win contexts. If the return/merit is not considered/perceived as sufficient by stakeholders, data will probably not be transferred. The idea of spontaneous willingness for free dissemination of these data is clearly naïve.

To sum up, these aspects (insufficient harmonization in the definition and data recording of the health KPIs, the issue sensitivity and high confidentiality level for this kind of information and also the distrust on the potential misuse of this information) are some of the reasons why farmers are reluctant to share recorded data on fish diseases.

The increasing use of big data, how this big data is generated and the debate about the availability of public information vs privacy of the information and data protection in the global world is affecting many different aspects of the society. This debate is also involving the question about where to draw the line and where to place the appropriate balance between what should be considered as public information and what can be considered as data with restricted access or private information. On the other side, it is also very clear that transparency is a clear advantage and one of the bases in the certification systems. This is the reason why it is so necessary to reach an operational balance between transparency and adequate communicative actions.

Like other aspects of fish farming activity (feeds, welfare), data and information from diseases and health aspects require careful and specialized processing by experts on the field but also by communication experts. There are no doubts that perception of the fish diseases amongst aquaculture experts is much more different than perception amongst consumers or public in general. Consequently, some kind of 'translation' mechanisms are necessary, in the same way that doctors have specific communicative strategies with patients and relatives.

In many cases, independent health experts such as private practitioners and fish health consultants play a relevant role as a bridge and filter this information. They know and work (but do not own) with these data and normally have or generate aggregated information from the same farms or other farms so they can have a much more general idea of the different aspects involved in the health of the farm before they develop recommendations for improvement. At the same time, they are absolutely aware of the importance or the sensitivity of the information and know what can be transferred, when and in which way should be addressed. Some aspects of the epidemiological information are particularly important and sensitive:

a) Geographical identification: nobody wants to see a map with a 'black flag with skull and bones' attached to the place of his/her farm. As it is said, 'hate the sin, not the sinner'. Sometimes, important information on epidemiological results is connected with geographical locations and sometimes this information is revealed even unintentionally, with undesired consequences. This can affect an area but also a region and even a country. Mediterranean fish farming is mostly concentrated in large facilities in the same site, that in some cases makes it really easy to identify a farm simply with few data.

For example, specific sites in Canary Islands, as a particular place for European seabass farming outside the main Mediterranean are very easy to be identified if data are not formatted appropriately. Also, countries or regions with a single farm with a specific activity (hatchery,

nursery...) can also be easily identified. Companies based in small countries, in islands and in particular places can be sometimes identified even in aggregated results when main categories are identified in the database.

b) Commercial relationships between farms and companies should also be considered. Although there is a general trend of vertical integration in the bigger farms or companies, eggs, larvae or juvenile trade from external hatcheries is still a very common trend in Mediterranean finfish aquaculture. Inadequate dissemination of information about health problems in these facilities may seriously harm their prestige.

As all these considerations should be taken into account, some examples of initiatives on collection and management of information of certain terrestrial animal diseases were explored. One of the systems also used for some human diseases are the **sentinel networks**.

Sentinel networks in human medicine

Many different and specific sentinel surveillance programs are already developed under WHO or other international health organisation coverage. Human influenza is maybe one of the diseases most widely known with a wide sentinel surveillance by [WHO](#) and the EC ([ECDC](#)).

Sentinel networks in veterinary medicine

Amongst terrestrial animal diseases, some similar initiatives, adapted to the specific characteristics of terrestrial farming have also been launched (e.g [the Ontario Animal Health Network Swine Project](#)). Sentinel surveillance can also be designed from a very local system, to national system and also can be expanded to the international level. The concept of sentinel surveillance networks is based on the selection or experienced and highly-qualified experts on the basis of a high probability of confronting cases of the selected disease/diseases. Another characteristic of the sentinel networks is a high and standardised quality of data collection. This specific data collection significantly complements and evaluates the information obtained or received by passive surveillance. These sentinel surveillance system not only can offer robust and trustable information about the current status of the disease, but also can identify significant changes in the trend of some diseases and the emergence of new diseases, providing a fast and efficient system for the industry.

These sentinel surveillance systems should also be based in the robustness and trustworthiness of the system and this implies the development of a system with a high level of data protection and confidentiality management but also a system of quality assessment. Another characteristic of these sentinel surveillance networks is the willingness to participate and safeguard the high level of trustworthiness of the system.

It also should be stressed that in Spain, a specific initiative on aquatic animal health was launched several years ago. This initiative was based on the so-called AD SG concept (*Asociaciones de defensa sanitaria ganadera* / Health-protection farming associations). This concept arose in the 90's from farmer associations with the common aim to improve the health status of their farms and with common disease prevention programs. These AD SG were particularly helpful for small-medium farms that had low or very low technological animal health and welfare basis and were also very useful to control certain diseases in Spain such as brucellosis or African swine fever. These associations were also recognized by the Spanish

Animal Health authorities and worked together with administration officers in mitigating diseases under official control.

Few years later, this idea was also developed for aquaculture and some continental aquaculture ADS were created in Spain mainly regarding trout health control. The idea was successful and also other marine aquaculture ADS were set up. Nowadays, several marine ADS from the main gilthead seabream and European seabass (Valencia, Murcia, Andalucía and Canary Islands) production areas are associated in a federation (FEADSA).

Frequent contact all year round between ADS/FEADSA health experts is very important, as they share general and updated information of the evolution of the diseases of importance for the fish farming sector. Furthermore, these experts are also in close contact with fish health representatives from the fish farms and companies, allowing a fluent transmission of data using a specific information channel, with quality information amongst the right people and also maintaining a high level of discretion in the information.

Particularly for gilthead seabream and European seabass disease surveillance, PerformFISH through its benchmarking system designed in WP7 offers a unique opportunity to develop sentinel system in the future. This system could be based on an adaptation of the KPIs scoring and benchmarking system, securing a high level of confidentiality, in a participatory initiative based on the willingness of the participants. Experts can be easily identified from the previous efforts made by Niccolò Vendramin and EURL and also both PerformFISH WP3 and MedAID WP4, as the people and institutions involved in the respective WPs of both projects represent most of the top experts on gilthead seabream and European seabass diseases and health management. This idea of sentinel group on European seabass and gilthead seabream health and diseases assessment perfectly fits with the EU Blue Growth Strategy and it could also be implemented through specific strategies for the Mediterranean Sea basin or through other platforms such as COST actions.

4. Diseases and disease management: requirements, hurdles and potential improvements for the future

Independently to the gilthead seabream and European seabass disease official or non-official monitoring strategies that can be established in the EU or in each country, there are several basic and major problems related to disease management that should be sorted out immediately.

These problems are as follows:

1) Endemic diseases:

As it is explained before, in the Mediterranean context, most of the diseases can be considered as endemic and particularly holoendemic. Although the occurrence and the impact of these diseases can vary in each area depending on several factors such as water temperature, water currents and presence of reservoirs, most of the pathogens are already present in most areas of the Mediterranean sea. In this scenario, disease transmission mainly takes place when gilthead seabream or European seabass fry are stocked in the cages and usually the period with higher mortality and morbidity is the period just after stocking. This means that gilthead seabream and

European seabass reared in nursery and pre-ongrowing sites normally come with reasonably high health standards and become infected when they are transferred to harder conditions, where these fish are exposed to a higher infective pressure from the neighbour cages and the wild fish populations around the cages. Thus, the role of disease transmission associated to juvenile transport has a lower relevance in the Mediterranean fish farming context compared with other finfish aquaculture farming activities.

2) Production in cages:

Gilthead seabream and European seabass are mainly produced using offshore floating sea cages. This production system presents a low isolation level from the environment and has a particularly high level of exposure to pathogens and perpetuation of the diseases in the cage. Therefore, this system presents risks in the development of diseases and presents severe limitations in the possibilities of controlling these diseases. According to that, from the sanitary point of view, gilthead seabream and European seabass production in cages raises clear limitations in the escalation into more intensive production technology.

3) Mixed age-class batch replacement system vs isolated single age-class batch replacement system

In a large number of farms, batches with different age-class stocks are placed together or in the vicinity with other stocks. After harvesting, cages are re-stocked again with new fry. These newly introduced fry are not only exposed to environmental pathogens, but also to pathogens from the neighbouring cages that contains bigger fish that normally are carriers of pathogens and parasites that very easily are transferred to the new fry. In other farms, cages are grouped according to age-classes, although this strategy normally requires bigger size of the farm maritime concession or to manage different maritime concessions. This strategy also requires the availability of bigger fry batches to fill several cages at the same time. This requirement for bigger batches of gilthead seabream and European seabass fry also requires relevant reconfiguration of the production in the hatcheries, nurseries and pre-on-growing facilities.

4) Fallowing / site rotation:

For cage production, site rotation and fallowing has been demonstrated as very efficient strategies to reduce the load of pathogens in a certain area. However, this method requires a wide availability of mooring places and administrative concessions. In the Mediterranean, due the co-existence of many human activities (tourism, transport, fisheries, commercial routes, protected areas,..) the coastal area for aquaculture activities is very limited. Sites on fallowing or not in production are in many cases an unaffordable luxury for the companies and farms. It should also be stressed that the reduced availability of maritime space for cage farming may press towards the concentration of farms in a certain area. This concept of ‘specific areas for aquaculture’ (IUCN, 2009) usually takes into account governance, social, economic and environmental aspects, but rarely takes also into account disease management requirements.

5) All in-all out systems

For animal production with large numbers of fish reared in the same site (hundreds of thousands to million individuals) under intensive production regimes, all in-all-out production systems using sanitary-certified fry in isolated premises with high hygiene and disinfection is undoubtedly the preferable system. In cages, single-age cage grouping seems to be a good

alternative to implement this system, although isolation levels are too low. Flow-through inland farms and mainly recirculation systems offer better opportunities to fulfill all in-all-out requirements.

5. Diseases and public information

Diseases can also be considered as a very relevant topic to the general public opinion. Amongst the different main perceptions and concerns of the public about aquaculture (Schlag, 2010), health-related issues such as diseases present in the fish, transmission of the diseases to consumers, the use of chemical substances (antibiotics, antimicrobials) in disease treatments & prevention and welfare have a prevalent place. In general, consumers usually do not bear in mind that farmed animals can become sick unless they read, listen or watch information about specific disease problems. In these cases, such news broadcasting increases their awareness and consequently, increase their concern about that. This concern can be increased if the information is accompanied by images of moribund or dead fishes floating on the water surface or explicit images of fish with skin injuries or wounds and if the information is biased or presented in a sensationalist way. Although the disease is something intrinsic of any kind of animal production, consumers and public in general, do not normally perceive this fact as something disturbing, unless they receive overstimulation by overemphasized news. This situation has been recurrently seen in salmon industry in association with mortalities by ISA, *Aeromonas* or sea lice in the newspapers, radio, TV and social media. Like other diseases in finfish aquaculture, gilthead seabream and European seabass diseases can also be targeted by media in the same way and this information can cause a major effect on the consumer's perception if this information is not addressed in the right way. Therefore, a relevant point concerning disease and health policies in gilthead seabream and European seabass farming in Europe should be how to communicate diseases beyond the internal industry or scientific environment. The simple use of words such as viruses, bacteria or parasites, without appropriate clarifications and nuances by the communicator (journalist, representative of an institution or association, etc.) can lead to wrong or biased information. Even amongst specialists in animal health, mainly those with limited knowledge on aquatic animal diseases, some kind of information on *Vibrio* diseases can be related with other more well-known diseases such as cholera, caused by *Vibrio cholerae*, or problems by *Vibrio parahaemolyticus* as food-borne disease, or even the assimilation of aquatic *Mycobacteria* to the much pathogenic causing problems in humans. Thus, it is paramount that diseases and disease-related information is communicated to the administrators and the public by specialists, by using appropriate channels and appropriate language.

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