Land based on-growing of Atlantic cod (*Gadus morhua*) and salmon (*Salmo salar)* using Recirculation Aquaculture Systems in the Basque Country: contributions to scientific understanding of economic feasibility, environmental sustainability, and societal acceptability

By

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Dr. Diego Mendiola

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Doctoral programme

Marine Environment and Resources

Department

Zoology and Animal Cell Biology

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Year 2017

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Universidad del País Vasco
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Thinking out loud….. When your mind goes faster than your body you feel that you will not be able to do anything else, you feel that the world has beaten you. However, it is when your mind is stronger when you know that you do not have any limit. The psychological strengthen wins over everything. Your force of will overcomes any boundary in your road. Your life may completely change in few hours but you need to be strong enough to face it. Do not put limits to yourself and rely on people that are worth; they will never leave you fall.

I have gone through all this sensations during my PhD studies. But I am here, presenting my work. It has taken longer than expected; the recovery and gaining confidence with me has been frustrating at the beginning, though afterwards and encouraging at the end.

Altuan hitzegiten…Zure burua, gorputza baino azkarrago doanean ezertarako kapaz zarela sentitzen duzu, munduak irabazi zaituela sentiarazten duzu. Aldi berean, zure burua ondo dagoenean inolako mugarik ez duzula badakizu. Indar psikologikoak beste edozerren gainetik irabazten du. Zure indarkeriak edozein muga pasa dezake. Baliteke zure bizitza derepentean aldatzea baina gogorra izan behar duzu aurre egiteko. Ez ezaiozu zure buruari mugarik jarri eta benetan lagunduko zaituen pertsonengan konfiatu, ez zaituzte jauzten utziko.

Guzti hau sentitu dut nire barnean doktoretzaren prozesuan. Baina hemen nago, nire lana aurkezten. Uste nuen denbora baino gehiago eraman dit. Hobekuntza eta nireganako konfiantza berreskuratzea frustragarria izan zen hasieran, oso gogorra aurrerantzean eta pozgarria bukaeran.

Hablando en alto….. Cuando tu mente va más rápido que tu cuerpo sientes que no podrás hacer nada más, sientes que el mundo te ha derrotado. Sin embargo, es cuando tu mente es más fuerte cuando no tienes límites. La fuerza psicológica gana sobre todo lo demás. Tú fuerza de voluntad sobrepasa cualquier barrera en tu camino. Puede que tu vida cambie completamente en pocas horas pero tienes que ser fuerte para afrontarlo. No te pongas límites y ayúdate de la gente que merece la pena, nunca te dejaran caer.

He sentido esas sensaciones durante mi doctorado. Pero estoy aquí, presentando mi trabajo. Ha llevado más tiempo del pensado. La recuperación y la adquisición de confianza en mí misma ha sido frustrante al principio, muy duro más adelante y alentador al final.

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Acronyms and abbreviations

AD: Abiotic Depletion

ANCOVA: Analysis of covariance

ANOVA: Analysis of variance

AOAC: Official Methods of Analysis of AOAC International

AP: Acidification Potential

APROMAR: Asociación Empresarial de Productores de Cultivos Marinos

BAP: Best Aquaculture Practices

BC: Before Christ

BFT: Biofloc Technology

BP: Barometric Pressure

Br: Bromide

CF: Condition Factor

CIWF: Compassion in World Farming

Cl: Chlorite

CO_2: Carbon Dioxide

COM: European Commision

DAPA: Departamento de Agricultura, Pesca y Alimentación del Gobierno Vasco

DHA: Docosahexaenoic acid

DO: Dissolved Oxygen

DPD: Total Chlorine method

EAS: Europeam Aquaculture Society

EC: European Commission

EEA: European Environment Agency

EIA: Energy Information Administration

EJ: Eusko Jaurlaritza

e.g.: For example

EP: Eutrophication Potential

EPA: United States Environmental Protection Agency

EPA: Eicosapentaenoic acid

EU: European Union

FAO: Food and Agriculture Organization of the United Nations

FCR: Food Conversion Ratio

FTS: Flow-through system

FU: Functional Unit

g: gram

G: Growth

GDP: Gross Domestic Product

GSSI: Global Sustainable Seafood Initiative

GV: Gobierno Vasco

GWP: Global Warming Potential

HSI: Hepatosomatic Index

ICES: International Council for Exploration of the Seas

i.e.: Id est

IMTA: Integrated Multi-trophic aquaculture

ISA: Infectious Salmon Anemia

ISO: International Organization for Standardization

K: Relative rate of growth

kg: kilo-gram

kWh: kilowatt-hour

L: Instantaneous body length growth

LCA: Life Cycle Assessment

LCI: Life Cycle Inventory

LCIA: Life Cycle Inventory Assessment

LOX: Liquid Oxygen

M: Mortality

MAGRAMA: Ministerio de Agricultura y Pesca, Alimentación y Medio Ambiente

MBTH: Formaldehyde MBTH method

MIB: 2-methylisoborneol

MW: Megawatt

N: number of fish

NASCO: The North Atlantic Salmon Conservation Organization

NO_2: Nitrite

NO_3: Nitrate

NOAA: National Oceanic and Atmospheric Administration

NR: Non-renewable energy

O_2: Oxygen

OPP: Organización Productores Piscicultores

***p*:** p-value

PNR: Point of No Return

R: Renewable energy

RAS: Recirculating Aquaculture System

ROI: Return On Investment

SC: Scenario

SD: Standard Deviation

SSA: Specific Surface Area

SST: Seawater Surface Temperature

SWOT: Strengthens-Weaknesses-Opportunities-Threats

T: Temperature

t: tons

TAN: Total Ammonia

TEL: Threshold Effects Level

TGP: Total Gras Pressure

UK: United Kingdom

UNE: Una Norma Española

US: United States

USDA: United States Department of Agriculture

UV: Ultraviolet

W: Instantaneous body weight growth

Laburpena

Arrainen hazkuntza kontuan harturik, abeltzantza da bai klima aldaketa, bai ur eta lur errekurtsoen faltaren arrazoirik garrantzitsuena. Gainera, industria honen beharrezko intentsifikazioak, ingurumenarekiko kezka sortarazten du globalki. Tesi honen ***Sarrera orokorrean*** azaltzen den moduan, akuikulturan, errezirkulazio sistemak dira produkzio mailan arrainen hazkuntzak sorturiko ingurugiroaren inpaktuen irtenbide. Sistema hauek ingurumen kontrolatu batean oinarriturik daude, sistema bakoitza desberdina da eta uraren tratamenturako erabilitako teknologia faktore desberdinen arabera (erabilitako ura, hazitako espeziea edota pentsuaren osagaiak) alda daiteke.

Aurkezturiko tesi honek errezirkulazio sistemen bitartez Euskal Herrian bakailao atlantikoa (*Gadus morhua*) eta izokin atlantikoa (*Salmo salar*) hazi ahalko liratekeen ikertzen ditu. Ikerketa lau ikuspuntu desberdinetatik egiten da: teknologia, ekonomia, ingurugiro eta gizarte edo kontsumitzaileen ikuspuntuetatik. Ingurumenarekin parekaturik, errezirkulazio sistemen teknologiaren bideragarritasuna aztertzen da; tokiko merkatuetan arrainak produzitzeak dakartzan koste/etekinak aztertzen dira; ingurumenean eragindako inpaktuak aztertzen dira eta tokiko kontsumitzaileen eta arraien inguruko adituen partetik produzituriko arraiaren onarpena ikertzen da. Gainera, metodologia berri bat aurkezten da errezirkulazio sistemek ingurugiroan duten inpaktua aztertzeko balio duena. Azkenik, teknologia honek energiaren erabileraren eta uraren tratamenduaren inguruan aurkezten dituen arazo haundienak eta etorkizuneko erronkak bistaratzen dira. Tesi honen helburu espezifikoak bost ezarpen desberdinetan aurkeztu dira.

Lehenengo eta ***Bigarren Ezarpenetan*** bakailao eta izokin atlantikoak hazteko bi esperimentu aurkezten dira. Alde batetik, ***Lehenengo ezarpenak***, bakailao atlantikoa errezirkulazio sistemen bitartez hazi daitekeen jakiteko, ekonomikoki aukera desberdinak aurkezten dituzten egoerak eta biologian eragina izan dezaketen faktoreak ikertzen dituen bideragarritasun azterketa bat aurkezten ditu. Horretarako, bi mila eta bostehun bakailau hazi ziren bi tenperatura desberdinetan (bata kontrolpean eta bestea itsasoko uraren tenperatura jarraituz) esperimentu eskalako bi errezirkulazio sistemetan Euskal Herriko kostaldean. Esperimentuak 430 egun iraun zituen. Diferentzia estadistikoak aurkitu ziren bi tenperatura desbedinetako arraien biziraupenetan nahiz eta ez ziren diferentzia estadistikoak aurkitu udazken edota negu denboraldietan. Hazkunde espezifiko indizeak estadistikoki desberdinak gertatu ziren uda denboraldian zehar eta hazkuntza konpentsazioak ikusi ziren itsasoko uraren tenperatura jarraituz funtzionatzen zuen sisteman. Era berean, diferentzia estadistikoak aurkitu ziren arrainen gantz kantitatean uda partea eta geroko analizietan. Halaber, inolako diferentziarik ez zen ikusi esperimentuan hazitako eta naturatik harturiko arrainen arteko zentzumen azterketan. Elektrizitatea izan zen ekonomikoki gastu gehien sortu zuen azterturiko faktorea. Esperimentu hau izan da errezirkulazio sistemen bitartez bakailaoa hazteko Espainiako iparraldean egindako lehen proba. Eskualdean posiblea dela teknikoki bakailaoa haztea ikustarazten du, aurretiaz egindako beste esperimentu batzuetan lorturiko hazkuntza balioak baliokidetzen ditu, bakailoa hazteko ur tenperaturaren kudeaketaren erabilgarritasuna proposatzen du eta eskualdean aktibitate komertziala lortzeko ekonomikoki klabeak diren parametro eta limiteak zehazten ditu.

Bestalde, ***Bigarren ezarpenean***, izokin atlantikoaren konpentsazio hazkuntza aztertzen da bertoko itsas tenperatura profila erabiliz eta baita produzitutako produktuaren kontsumitzaileen onarpena eta produktua erosteko intentzioa ebaluazio hedoniko baten bitartez. Horretarako, mila eta bostehun izokin hazi ziren 497 egun iraun zituen esperimentuan zehar bi errezirkulazio sistemetan. Hazkuntza indizeak altuagoak izan ziren bi tenperatura erregimenetan uda partean eta nolabaiteko konpentsazio hazkuntza ikusi zen itsasoko uraren tenperaturaren menpean lan egin zuen sisteman. Aitzitik, ez zen inolako diferentziarik ikusi zentzumen produktuaren zentzumen analisian esperimentuan hazitako izokin eta merkatuan erositako produktuaren artean (Dinamarkan errezirkulazio sistemen bitartez produzitutako izokina). Produktuarekiko kontsumitzaileen onarpen maila altuak eta erosteko intentzioak posible egiten dute eskualdean produzitutako izokinaren salmenta. Aldi berean, esperimentu hau da Espainiako iparraldean inoiz egin den izokina errezirkulazio sistemen bitartez produzitzeko azterketa.

Errezirkulazio sistemak arraina produziteko sistema intentsiboak dira, ur eta lur gutxiago erabiltzen dutenak. Hala ere, beharrezkoa duten energia kantitate haundiak, gehienbat erregai fosiletan oinarrituak direnak, koste operazionalak eta ingurugiroan inpaktuak handitzen dituzte, beraien hedapenerako eragozpen bat izanik. Hala, ***Hirugarren*** *eta* ***Laugarren ezarpenek*** energiaren garrantzia ikertzen dute sakonean, honen ebaluazio eraginkorrago bat egitera laguntzen eta energia aurrezteko moduak aurkezten dituzte. Energiaren erabilira errezirkulazio sistemetan zeharka ikertu edota argitalpen gutxitan aipatu da. Horrez gain, honen garrantzia eta inpaktuak ez dira aztertuak izan. Gutxi balitz, ekonomikoki eta ekologikoki produkzio eraginkor eta pairagarri bat lortzeko konpromiso bat bilatu beharko litzateke uraren erabilera eta, energiaren kontsumoa eta produktibitatearen artean.

Hala, Hirugarren ezarpenak, energiaren erabileraren inguruan orainarte argitaraturiko azterketak aztertzen ditu. Gainera, industriarentzako baliagarria izateko asmoarekin produzitzaileei zuzenduriko galdetegi bat egin zen. Diseinu eraginkor eta energia gutxiagoren menpeko bat aurkezten du optimizaturko prozesuak bateraturik, integraturiko sistema definitzen du eta baita ekipo desberdinen aukera. Ondorio garrantzitsuenak hauek izan ziren: energia berriztagarriak fosil erregaiak baino koste-eraginkorragoak direla, industriarentzako energia ez dela inolako kezka eta energia berriztagarriek errezirkulazio sistemen industrian pontentziala dutela. Honen arabera, ***Laugarren ezarpenak*** bi metodologiaren konbinazioa aurkezten du: produktuen bizitza ziklo ebaluazioa eta auditoria energetikoak. Honen helburuak hauek izanik: errerzirkulazio sistemen ingurumen errendimendua hobetzea eta energia kontsumoak dakartzan ingurumen eta efektu ekonomikoak identifikatzea kostuak murrizteko. Proposaturiko metodologia Lehenengo ezarpenean aurkezturiko esperimentuan probatu zen. Nahiz eta sistemak batazbesteko 29.40 kWh/kg arrain erabili zituen, energia kontsumoak bariabilitate haundia izan zuen denboraldien arabera, maximo (40.57 kWh/kg arrain) eta minimo (18.43 kWh/kg arrain) batzuk aurkeztuz. Sistemako kontsumitzaile haundiena ura hozteko beharreko bonba izan zen, bonba printzipal eta bigarren mailakoak jarraiturik. Auditoria energetikoa lagungarria gertatu zen kontsumitzaileen identifikazioan eta erregistraturiko datuek produktuen bizitza ziklo ebaluazio oso eta zehatzago bat egiten balio izan zuten. Erregai fosiletan oinarrituriko elektrizitatea izan zen ingurumenean inpaktu gehien sortzen zuen faktorea. Kontsumo aldakorra aurkeztu zuen uraren tenperaturaren arabera; temperatura da energiaren kontsumoa zuzentzen duen parametroa. Produktuen bizitza zikloak eta auditoria energetikoaren konbinazioa oso tresna erabilgarria dela ikusi zen energia gutxiko sistema produktibo eta eraginkorrago bat diseinatzeko. Aldi berean, gobernantza eta erabakiak hartzeko hastapena azkartzen ditu, denboran oinarrituriko energia kontsumoaren fluktuazioa produktuaren bizi-ziklo osoan zehar kontuan harturik.

Hala, ***Bostgarren ezarpenean***, errezirkulazio sistemen arazo garrantzitsuenak aztertu ziren etorkizunean produkzio zuzendariei irtenbide hobeagoak emateko; beti ere industrian hobetu beharreko esparruak eta etorkizuneko erronkak identifikatuz. Errezirkulazio sistemetan oinarrituriko enpresak, ikertzaileak, sistema disenatzaileak eta aholkulariak elkarrizketatu ziren banan banan, sistemen ulermen orokorra eta zein garapenek lagunduko luketen ikertzeko. Jasotako erantzunek eta geroko analiziek produzitzaileen parte-hartze pobrea, informazioa elkarbanatzeko oztopoa eta talde desberdinen arteko komunikazio falta identifikatu zituzten barrera moduan. Identifikaturiko arazorik nagusienak: sistemen diseinu pobrea eta beraien kudeaketa eskasa. Nabarmenduriko lehentasunak sistemen errendimendua hobetu beharra eta ikerketa gehiago uraren tratamendurako erabilitako tresnen konbinazioan egoera espezifikoetarako. Honetaz aparte, espezialisten plataforma bat sortzea gomendatzen da, nun errezirkulazio sistemen inguruan ezagutza elkarbanatzen den hezkuntza programa sakon eta beraizgarriekin batera.

Azkenik, ***Eztabaida orokorrean***, Ekarpen desperdinetan izandako emaitzak ikuspuntu integratzaile batetik analizatu dira aurretik zehaztutako helburuei erantzunez. Eztabaida lau ikuspegi desberdinetan banatu da eta bestelako ikuspegietatik (enpresa zuzendaria edota arrain kontsumitzailea) sortutako galderak erantzuten dira. Orokorrean, tesi honetan proposatutako analisi zehatzak, eskualdean errezirkulazio sistema batean oinarrituriko akuikultura konpainia bat sortzeko beharrezkoak diren pausoak aurkezten ditu. Teknologi, ekonomia, ingurugiro eta kontsumitzaileen aspektuak biltzen ditu bakailao eta izokinaren produkzioaren inguruan, bakoitzarentzako azterketa luze eta zehatza eginez. Teknikoki, esperimentu mailan izan bada ere, arrainen hazkuntzarako tenperatura estrategiak probatzen dira. Hala ere, ikerkuntza zabalagoa egin beharko litzateke inolako komertzial mailako ekimen bat proposatu aurretik, nun lan estrategia, hazitako espezie eta uraren tenperaturaren arteko oreka bat beharrezkoa den. Mundu mailan teknologia honek duen garrantziak eta ingurugiroaren ongizaterako kontsumitzaileen gero eta kontzientzia haundiagoak, beharrezkoa egiten du errezirkulazio sistemen aspektu guztien hobekuntza gero eta enpresa gehiago sortu eta akuikultura jasangarri eta ingurugiro lagungarria izan daiten.

Summary

Animal farming systems, including fish farming, are regarded to be a major cause of problems such as resource (water and land) depletion and climate change. Moreover, their required intensification represents a relevant cause of environmental concern at global level. As it is remarked in the ***General Introduction*** of this thesis, in aquaculture, the option for reducing the environmental footprint of aquatic animal production and mitigating many of the impacts associated with traditional commercial fish culture technologies (i.e. net pens, ponds, flow-through systems) is Recirculating Aquaculture System (RAS) technology. RASs are based on highly controlled environments. Each system is different and the technology used in the water treatment loop may differ depending on several factors (e.g. water, species reared and feed’s ingredients).

The present work studies the feasibility of using RAS to rear cold water species such as Atlantic cod (*Gadus morhua*) and Atlantic salmon (*Salmo salar*) in the Basque coastal area. The study is made from four different perspectives: technical, economic, environmental and societal. The viability of the technology is tested coupled with the surrounding environment; the cost/benefits of producing fish in the local market are analyzed; the environmental performance and the created impacts are studied; and the final product´s acceptability is studied among local consumers and seafood experts. Moreover, a new environmental assessment is presented and detailed knowledge of the main issues and future challenges are obtained regarding energy use and water treatment technology.

Contribution 1 and ***2*** displayed two experimental approaches to rear Atlantic cod and Atlantic salmon, respectively. In one hand, ***Contribution 1*** presented a feasibility study to analyze the different economic scenarios and biological factors that can influence the business potential of growing Atlantic cod in RAS. For that, 2,500 cod individuals were reared at two different thermal regimes (i.e. controlled and natural range, respectively) through 2 pilot RAS set up in the Basque region (Northern Spain). The experiment lasted 430 days. Statistical differences were found in survival between different thermal regimes but no significant differences were detected within the fall or winter seasons. Daily specific growth rates were significantly different during the summer season with some compensatory growth patterns being observed in the natural thermal regime set up. Likewise, statistical significances were found between the fat contents from both temperatures after the summer period. Conversely, no significant differences were observed at sensorial level between the samples obtained within our pilot experiment and commercial samples from wild origin. Electricity use was found to be one of the most significant economic costs to be considered. The study represented the first technical feasibility attempt on cod in land-based aquaculture from the north of Spain and demonstrated the technical feasibility to produce on-land based cod in the region, the equivalence of growth patterns with previous studies, the usefulness of the proposed thermal regime management as a tool for this species production, and the key economic parameters and thresholds for a potential feasible commercial activity in the region.

In the other hand, ***Contribution 2*** studied the compensatory growth of Atlantic salmon using local seawater temperatures, and consumers’ final product acceptance and purchasing intention through a hedonic evaluation. For that, 1,500 salmon individuals were grown for 497 days at two different thermal regimes in two pilot-scale RAS units. Growth rates were significantly different for both temperature regimes during the second summer season with some compensatory growth patterns being observed along the timing of the natural thermal regime set up. Conversely, no significant differences were observed at sensorial level between the fillet samples obtained in this study and commercially grown RAS salmon from Denmark. Consumer level of acceptance and product purchasing intention reflected the possibility of marketing RAS grown salmon in the local markets. Likewise, this study referred the first technical attempt on salmon land-based aquaculture from Northern Spain.

RAS are intensive fish production systems, with reduced use of water and land. Nevertheless, their high energy requirement is a drawback, which increases both operational costs and the potential impacts created by the use of fossil fuels. Thus, ***Contribution 3*** and ***4*** aimed to study more in detail the importance of the energy use, contribute to its more efficient assessment and provide energy saving measures. Energy use in RAS has been studied indirectly and/or mentioned in several publications. Nevertheless, its importance and impacts have not been studied. Herein, in aiming to achieve economic and environmentally sustainable production a compromise has to be found between water use, waste discharge, energy consumption and productivity.

Thus, ***Contribution 3*** discussed the published studies about energy use and RAS designs efficiencies. Moreover, with the aim of making an industry base study a questionnaire about the energy use in commercial scale RAS was conducted. The design of more efficient and less energy dependent RAS was presented, including optimized unit processes, system integration and equipment selection. The main conclusions were that fossil based fuels are less cost-effective than renewable energies; energy is of little concern for the majority of the industry, and renewable energies are of potential use in RAS. In accordance, ***Contribution 4*** proposed a combination of two methods (i.e. Life Cycle Assessment (LCA) with energy audits) to: improve environmental performance of RAS, identify energy consumption and thus, its environmental and monetary effects in order to seek cost reduction. The proposed methodology was proved with the case study presented in Contribution 1. Although the system required an average of 29.40 kWh/kg fish for successful system operation, the energy consumption varied by season presenting maximum and minimum periods of 40.57 and 18.43 kWh/kg fish, respectively. Main consumers included the heat pump, followed by the main and secondary pumps, respectively. Energy audit’s results showed the success in identifying the devices that consumed the largest amount of energy, and recorded data served to feed the Life Cycle Inventory and perform a more complete and precise LCA. Fossil fuel based on-farm electricity for the on-growing of fish was shown to be the most environmentally unfriendly input; it was the major impact producer in the assessed impact categories. It showed a temporal variability depending on the water temperature, which resulted to be the main factor linked to the energy use. This aided performing a precise assessment including system-specific scenarios. The combination of LCA and on-farm energy audit represented a useful tool to secure a more complete assessment with a periodic assessment to design a less energy intensive, profitable and sustainable system; likewise, it increases the speed and transparency of governance and decision-making, taking into account the time-based fluctuation of the energy consumption throughout the production cycle.

Therefore, in ***Contribution 5***, main issues for RAS were analyzed, in order to lead to better solutions for future managers, identifying possible areas for improvements and future challenges for the industry. RAS-based production companies, researchers, system suppliers and consultants were interviewed separately, in order to gain an overall understanding of those systems and what developments could assist, in a positive way. Answers and subsequent analysis identified as significant barriers: poor participation by the producers; a disincentive on sharing information; and a lack of communication between different parties. The main issues identified were: poor designs of the systems and their poor management. Highlighted key priorities are the necessity to improve equipment performance and further work on the best combinations of devices for each particular situation. Additional recommendations are for a specialized platform, to share knowledge on RAS, together with a more in depth and distinctive education programme.

Finally, in the ***General discussion***, the results obtained in the different Contributions are analysed from an integrative point of view, in relation to the objective established for this thesis. The discussion is divided in the four different perspectives studied and questions raised are answered from different points of view (i.e. RAS manager and fish consumer). Overall, the tiered approach proposed in this thesis may be useful to set up a RAS company in the region. It comprehends technical, economic, environmental and social aspects of a potential cod or salmon production, making an extended analysis of each. Technically, a modulated temperature strategy is tested at an experimental scale. However, further investigations should be developed before proposing any commercial scale initiative regarding to the balance required between the working strategy, fish species reared and seawater temperature profile. The importance of this technology worldwide and consumer’s consciousness towards environment’s welfare makes necessary to improve every aspect of RAS to increase the number of companies pointing to a more sustainable fish industry.

General introduction

1. Intensive animal farming

Intensive animal farming is defined as crowding of animals closer together keeping them in larger groups to obtain faster growth or a major production per yield (FAO-CIWF 2015). Animal farming systems, including fish farming, are regarded to be a major cause of problems such as resource (water and land) depletion and climate change (Winther et al. 2009; Sonesson et al. 2010; Lesschen et al. 2011; Nijdam et al. 2012). Moreover, their required intensification (Steinfeld and Wassenaar 2007), as reported for other activities like agriculture or ranching (FAO 2006; Dumont et al. 2012) represents a relevant cause of environmental concern at global level (Pillay 1992; Muir et al. 1999; Tisdell 1999; Naylor et al. 2000; Frankic and Hershner 2003; Read and Fernandes 2003; Focardi et al. 2005; Crab et al. 2007; Sapkota et al. 2008).

At the early 1960s, at the time that any public policy was interested in overtaking the environmental problems related to the impacts of intensive agriculture, livestock farming and/or intensive aquaculture, some scientific movements (i.e. agroecology and industrial ecology) emerged against those industrialized activities (Wezel and Soldat 2009). Such disciplines encouraged production systems to minimize environmental impacts by re-inventing or adapting technologies and techniques to propose a more “environmentally friendly” farming systems. In such disciplines, intensive systems are productive and optimized, need few chemicals inputs, and are resource conserving while reusing wastes as inputs for another production processes (Frosch and Gallopoulos 1989; Frosch 1992; Gliessman 1997). Consequently, the concept of sustainable production emerged in 1992, at the United Nations Conference on Environment and Development. It was defined as the creation of goods and services: using processes and systems that are non-polluting; conserving of energy and natural resources; economically viable; safe and healthful for employees, communities and consumers; and socially and creatively rewarding for all working people (Veleva and Ellenbecker 2001).

1.1. Aquaculture

The practice of aquaculture (i.e. farming of aquatic animals, plants, fungi and other life forms of food) has sustained and enhanced human life for centuries, changing civilizations sedentary and farming organisms that created food surpluses to nurture, improve human condition and develop civilization since 2800 BC (Jesse and Casey 2006). Aquaculture and fishing are both important economic activities in Europe and worldwide. However, aquaculture is the fastest growing food production industry experiencing an unprecedented development in global animal production (Natale et al. 2013). Albeit, European policies struggle to solve causes and economic consequences for decline and impacts of marine fisheries (Pauly et al. 2003; Costello et al. 2008; Cheung et al. 2009; Natale et al. 2013; FAO 2012,2014; Natale et al. 2013), the rise of aquaculture represents more and more a contemporary phenomenon, in both production and value (McCausland et al. 2006; Asche et al. 2008; Duarte et al. 2009) (Fig. I.1). Global marine capture fisheries have been consistently harvesting between 80 and 90 million tonnes per year since the mid-1980s. In 2012 aquaculture provided almost 50% of all fish for human consumption and has been predicted to provide 62% by 2030 (Source: FAO 2014).

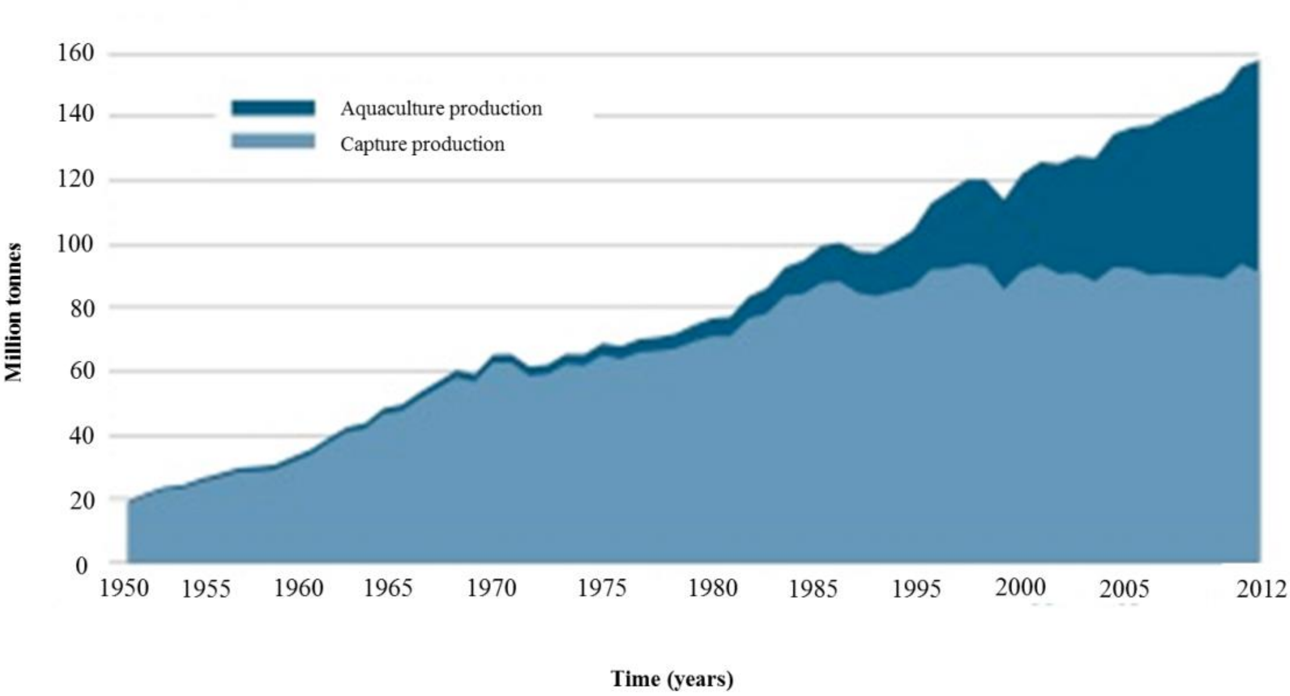


Fig. I.1. Aquaculture and capture fisheries production (million tons) between 1950 and 2012.

Aquaculture production has increased from 32.4 to 70.5 million tons between 2000 and 2013 (FAO 2014). It is the fastest growing food production sector. It has also the potential to generate local economic activity, and to bridge the growing gap between the demand and supply of aquatic products; provided always that natural resources are sustainably managed and the animal feeds industry reduces its reliance on wild fish (Olsen et al. 2008; Merino et al. 2012). Nevertheless, aquaculture's capacity to expand should be driven to reduce created environmental impacts (Naylor et al. 2009), intensifying its production in a sustainable way. This, coupled with the steady increase in the demand of fish (FAO 2014), has forced the aquaculture industry to look for tolerable solutions from environmental, societal and economic perspectives.

Recirculating Aquaculture Systems (RASs) are systems where water is re-used after undergoing treatment (Rosenthal et al. 1986). Ever since their inception they were highlighted as one possible way for reducing the environmental footprint of aquatic animal production and mitigating many of the impacts associated with traditional commercial fish culture technologies (i.e. net pens, ponds, flow-through systems) (Dalsgaard et al. 2013; Daniels 2014). Indeed, RAS, also known as "closed-containment systems", are currently the most preferred "green-technology" systems by companies (Martins et al. 2010; Klinger and Naylor 2012).

RAS became an alternative to industrial and environmentally friendly production systems. They are based on highly controlled environments (Dumont et al. 2012) and make possible correlations between both activities (Frosch 1992): outputs from fish farming as inputs for vegetables cultivation (i.e. towards zero creation of waste). However, despite their potential, RAS will only fall within the denomination of environmentally sustainable production systems if: (i) enhance animal health and production yield by adapting correct management practices and control measures; (ii) decrease inputs needed (i.e. water, electricity, feed); and (iii) decrease pollution by direct nitrogen removal and/or optimization of metabolic functioning (Altieri 2002; Dumont et al. 2012).

2. Recirculating Aquaculture Systems

2.1. General overview

RAS started to develop in the 70s based on sewage treatment plants. RAS are not simple systems; they are technology–biology interaction systems, requiring performance monitoring (Lekang 2007). They have benefited from continuous development (from the simplest path of water treatment until the most sophisticated process) (Muir 1982; Rosenthal 1993); nowadays, they are considered "high-tech" methods.

The design and engineering of RAS have been extensively studied over the years (e.g. Piedrahita et al. 1996; Van Rijn 1996; Cripps and Bergheim 2000; Summerfelt and Penne 2005; Eding et al. 2006; Summerfelt 2006; Morey 2009). Most of this research has been directed to: (I) improve particular devices, as well as individually the best performance (e.g. biofilters (Van Rijn 1996; Eding et al. 2006; Summerfelt 2006) and solids removals (Piedrahita et al. 1996; Cripps and Bergheim 2000; Summerfelt and Penne 2005); (II) compare different techniques (d'Orbcastel et al. 2009a; Pfeiffer et al. 2011a); and (III) design entire systems based on particular assumptions (Morey 2009). However, little has been done to describe potential risks (e.g. Hrubec et al. 1996) and issues whilst managing the system, and how all the components can be combined together. Most of the conclusions and studies relate to specific situations with a given species and growing parameters. Nevertheless, there are not 2 identical RAS and it is difficult to use one particular example to construct a good performance RAS (Piedrahita et al., 1996 cited this output of a workshop on Aquaculture Effluent Treatment Systems and Costs, held at Stirling University (June 1994)). The understanding of the system is one of the key factors in its management, as this requires interaction between engineering, and life organism biology and husbandry.

An example for a general RAS design is shown in Fig. I.2. As mentioned, each system is different and the technology used in the water treatment loop may differ. This will depend on: the water (i.e. marine or freshwater), species reared (i.e. cold- or warm-water species), and feed's ingredients (i.e. if the species is carnivorous or herbivorous), among other factors. Moreover, the most common operations/components within a RAS are: mechanical filtration (i.e. solid wastes removal) by filters (e.g. drum filters, sand filters); biofiltration (i.e. nitrogenous wastes removal) by biofilters (e.g. fluidized bed filters, trickling filters); disinfection by ozone and/or ultraviolet (i.e. UV); gas management (i.e. carbon dioxide (CO_2) removal and oxygenation) by additional oxygen supply and /or aeration; protein skimming by skimmers; and, denitrification (i.e. bioconversion of nitrate to nitrogen gas) by denitrification units. Regarding to the mentioned components, there are many different equipment manufacturers and styles, and their order within the water treatment loop can also vary (i.e. depending on the designer, species, and production volume).

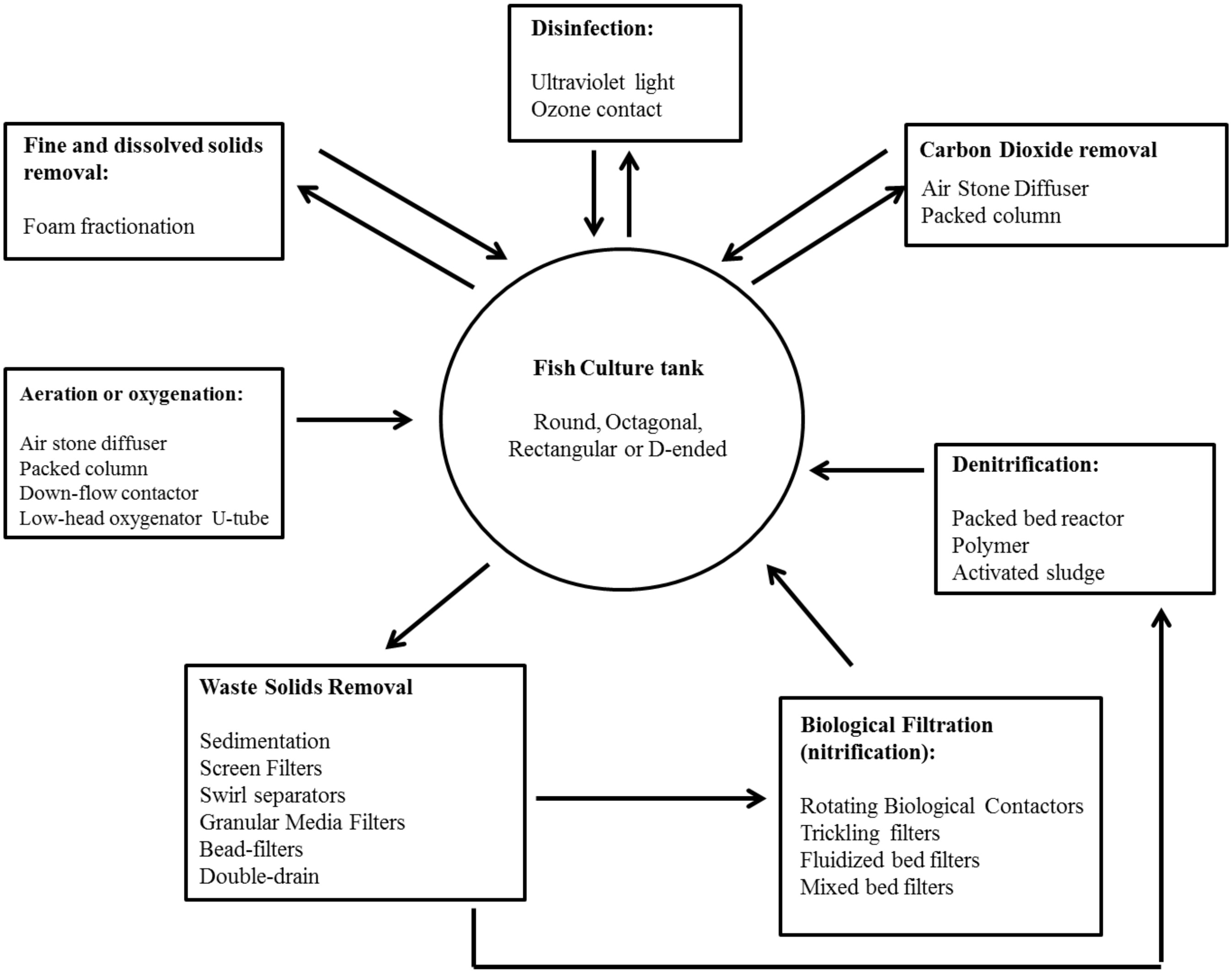


Fig. I.2. Required unit processes and some typical components used in recirculating aquaculture production systems (updated from Losordo et al. 1998).

2.2. Benefits and challenges

RASs present numerous advantages regarding fish production (Table I.1.). They can be located where the land is unsuitable for other food type industries (Zohar et al. 2005; Singer et al. 2008; Miller 2008). Likewise, they offer limited or non-interaction with the surrounding waters and natural habitats, eliminating so the negative interactions and environmental concerns reported from aquaculture activities (Labatut and Olivares 2004; Zohar et al. 2005; Buschmann et al. 2006). RAS allow greater control over the environmental and water quality parameters enabling optimal conditions for fish culture (Heinen et al. 1996), and less risk for the operator. A competitive advantage is achieved through many different advantages: fish wellbeing is maintained (d'Orbcastel et al. 2009a), both heat and water are conserved, and a consistent production schedule (varying from week, month planning) can be reached to control the harvest of fish when market demands, rather than when fish can be grown (van Gorder 1994). RAS are specially designed to be set up close to target niche-markets offering products, which are higher in fresh value (twice in price than the frozen one) and meat quality (Labatut and Olivares 2004; Timmons and Ebeling 2010). Furthermore, biosecurity levels are also higher when compared to conventional aquaculture system (i.e., flow-through systems (FTS), sea cages or ponds), decreasing the risk levels on disease threats.

ADVANTAGES	**REFERENCES**
Control over the environmental and water quality parameters	Heinen et al. 1996; Piedrahita 2003
Optimal conditions for fish culture-----animal welfare	d'Orbcastel et al. 2009a
Higher stocking densities	Tal et al. 2009
Possible location on-land unsuitable for other type of food productions	Zohar et al. 2005; Miller 2008; Singer et al. 2008
Location close to market	Masser et al. 1999; Labatut and Olivares 2004; Schneider et al. 2010
Consistent production schedule	van Gorder 1994
Decrease ecological impacts	
Water usage, decrease in water dependence	Chen et al. 2002; Blancheton 2000; Moss et al. 2001; Verdegem et al. 2006; Tal et al. 2009; Wik et al. 2009; Martins et al. 2010
Eutrophication potentia	Colt et al. 2008; Ayer and Tyedmers 2009; d'Orbcastel et al. 2009b; Eding et al. 2009
Better hygiene and disease management – Biosecurity	Summerfelt et al. 2001; Summerfelt et al. 2009a; Tal et al. 2009
Limited or non-interaction with surrounding environment	Buschmann et al. 2006
No escapees	Zohar et al. 2005
Ensure prevention of inclusion of pathogens guarantying chemical-free production	Badiola et al. 2014
Removed solids (rich in nitrogen and phosphorous) useful for agriculture	Cripps and Bergheim 2000; Piedrahita 2003; Marsh et al. 2005; Mirzoyan et al. 2010; Brown et al. 2011
DISADVANTAGES	
High investment	Schneider et al. 2006; Badiola et al. 2012
High operational costs (feed, labor, energy)	Sheperd and Bromage 1988; Aubin et al. 2006; 2009; Pelletier and Tyedmers 2007; Colt et al. 2008; Ellingsen et al. 2009; Jerbi et al. 2012
Technical skills required	Martins et al. 2010; Badiola et al. 2012
Energy intensive-----environmental and economic sustainability concern	Aubin et al. 2009; Ayer and Tyedmers 2009

Table I.1. A review of the main advantages and disadvantages of RAS.

However, the implementation of RAS involves many disadvantages (e.g. Liao and Mayo 1974; Sheperd and Bromage 1988; Blancheton 2000; Lekang 2007) and their management require specific technical skills, including some biological and engineering knowledge, as listed in Table I.1. (Badiola et al. 2012). The major constraints and limits of RAS are high economic costs (i.e. investment and operational costs) and the need of sophisticated technical skills for careful management (Wik et al. 2009; Martins et al. 2010). Constant internal pumping, new water intake and/or heating/cooling the water for production purposes lead to high electricity costs (Sheperd and Bromage 1988). Thus, energy use, may represent a sustainability concern from an environmental and/or economic perspective if correct decisions such as species selection, production dimensions and water pumping sites are not properly taken at the early stage of the RAS designing (Bostock et al. 2010; Martins et al. 2010; Dalsgaard et al. 2013).

2.3. Different initiatives for good environmental practices

The environmental impact of fish farming varies widely, depending on the species being farmed, the methods used, and where the farm is located. When good practices are used, it is possible to farm seafood in a way that has very little impact to the environment. Such operations limit habitat damage, disease, escapes of farmed fish, and the use of wild fish as feed. Eco-labelling and diverse accreditation initiatives (e.g. Seafood Watch recommendations) are arising within the food industry. A European-wide eco-labelling scheme was introduced by the European Commission (EC) in 1992 as part of its fifth and most recent Environmental Action Plan, the focus of which is also sustainability. The EC eco-labelling scheme aims to promote products with reduced environmental impacts throughout their life cycle and to provide consumers with better information about the environmental impact of products (Erskine and Collins 1997). The proliferation of voluntary certification and labelling schemes for environmentally and socially responsible production is often seen as driven by companies and consumer demand. The most popular and known certifications among the seafood industry are: Aquaculture Stewardship Council, Best Aquaculture Practices Certification (the responsible seafood choice) of Global Aquaculture Alliance, Certification of Canadian Farmed Seafood, Global Sustainable Seafood Initiative, and Whole Foods Market "Responsibly Farmed" Logo.

Also, there are organizations promoting the consumption of sustainable seafood and helping consumers to choose seafood that is fished or farmed in ways that have less impact on the environment. The most popular one is the Seafood Watch program from the Monterey bay Aquarium, which provides a traffic light colour-coded recommendation for each of the studied fish production company. Based on 10 environmental criteria, it indicates to the consumer if the product is avoidable (red), a good alternative (yellow) or the best choice (green). A specific report was undertaken for Global RAS (peer-reviewed), applicable to all species (Badiola et al. 2014) (available in https://www.seafoodwatch.org) which concluded a positive assessment. Overall, RAS was shown as a good production system with reduced environmental impacts associated with other aquaculture production systems (e.g., net pens, ponds, flow-through systems) and/or commercial aquaculture. However, energy use remained as one of the principal concerns. It was remarked that energy consumption should be the focus of further study and included as an environmental criteria of RAS.

2.4. Current research and future challenges

The basic RAS technology seems quite out-engineered. Nevertheless, their adoption in the future will be determined by the response of industry to the challenges they face. In the first instance, research and improvements, in terms of individual devices, should be directed towards commercial scale aquaculture, obtaining more reliable and useful data. Their operational systems will need to be better understood, in order to move towards a standardization of the industry. Moreover, in terms of improving their management and having more efficient and less failure prone systems, more specialized and highly capable people will need to be trained.

RAS are frequently not economically viable; "encouraging technology" is inevitable, but there must be an economic reason, in relation to an overall "market-need" oriented perspective of the system that ensures technically feasibility as a prerequisite to be economically viable. A good market or social study is needed, in order to meet with the actual demand, planning an affordable and realistic production goal. Thus, the first requirement is a reliable operation followed by low operating costs. Both conditions will aid recover more rapidly from the first investment: the first obtaining a stable production and, thus, profits; and the second providing a higher margin for the return.

Environmentally, research should be focused in: (I) more energy and cost-efficient systems; (II) a cradle-to-cradle approach system development; (III) reusing wastes for other purposes or product commodities; and (IV) use of alternative energy sources. Finally, a better marketable product will aid to improve the consumers' acceptability towards farmed fish.

In such context, there are currently various research areas:

- Use of renewable energy sources as part of the solution for RAS high energy requirements (i.e. high operational costs and ecological impacts created) (Aubin et al. 2006, 2009; Colt et al. 2008; d'Orbcastel et al. 2009b; Buck 2012).

- Better understanding of the interactions between rearing water and bio-accumulated compounds (i.e. geosmin and 2-methylisoborneol) (Tucker 2000; Howgate 2004) which are responsible of off-flavor fish in RAS.

- Closing RAS even further by the use of denitrification (i.e. nitrogen compounds such as nitrite and nitrate are reduced to elemental nitrogen) reactors and thus, improving management of wastewaters (i.e. usual organic carbon discharges are minimized as fish wastes are used to fuel denitrification). Several studies have been published with successful results (Gut et al. 2006; Klas et al. 2006; Tal et al. 2006, 2009) although the cost-effectiveness needs still greater understanding.

- In order to comply with water management and legislation, phosphate level control systems, which are currently accumulated within the systems (Martins et al. 2009), should be designed. Thus, the introduction of new compartments such as algal and for aquaponics production to (I) decrease environmental output, (II) valorize nutrients and detritivores taking advantage of produced byproducts such as carbon dioxide, and (III) generate secondly products to a major economical input may be an option. Additionally, Integrated multi-trophic aquaculture (IMTA) is also currently the most promising solution (Metaxa et al. 2006; Muangkeow et al. 2007).

2.5 Production volumes

There is not any official worldwide data regarding exact production volumes, monetary values, or number of RAS farms in operation. Indeed, the global RAS industry has been cited as doing a poor job of communicating information, as shown by various authors (Martins et al. 2010; Badiola et al. 2012; Dalsgaard et al. 2013) and international organizations (FAO 2012). However, according to reviewed publications (Sturrock et al. 2008; Bergheim et al. 2009; Martins et al. 2010; Badiola et al. 2012,2014; Dalsgaard et al. 2013; Murray et al. 2014) and some worldwide research made through personal communication and social networking by the author during the last 4 years, the number of RAS companies around the world keeps steadily increasing. The latest updates of the current number of RAS companies around the world are presented in the results section of this thesis (see Contribution 3, Fig. 3.1), being the US, by far, the country with more number of companies.

In the US, there are around 360 RAS companies currently in operation within the States of Florida (86), California (25), Virginia (19), North Carolina, Ohio, Wisconsin (16), Texas and Hawaii (15) (USDA 2014). The number of produced species represents a great variety (Table I.2).

Canada represents the 4th world´s largest production of Atlantic salmon (*Salmo salar*) (behind Norway, UK and Chile) (CAIA 2012); representing 67% of total aquaculture production volume in the country. While, most Atlantic salmon smolts are exclusively produced in RAS facilities before being sent to marine net-pen grow-out sites, several national RAS facilities for on-growing of salmon are currently being planned or constructed. Apart from this, several other species are also reared in Canada (Table I.2).

Europe has reflected a very relevant technical development and this has been reflected in somehow in the increase of production volumes in RAS (see Contribution 3, Table 3.2). Already, since the early 1900's, many fish hatcheries were operating as RAS (Blancheton 2000). In 1986, RAS production was concentrated in The Netherlands producing 300 t of rainbow trout, while in 2009 the industry was expanded all over the continent producing around 40,000 t/year marketable size fish and more than 147 million of fingerling head/year. Currently, the number of RAS companies is around 360 (Badiola et al. 2014) producing a wide variety of species (Table I.2) (Williot et al. 2001; Bergheim et al. 2009; d'Orbcastel et al. 2009c; Jokumsen and Svendsen 2010; Dalsgaard et al. 2013).

In South America, most of the aquaculture producer countries (i.e. Argentina, Colombia, Brazil) utilize production systems that experience high water exchange rates. The exception is Chile, where several RAS companies produce rainbow trout, abalone, turbot, Atlantic and coho salmon (Dempster 2014). In Asia, while RAS has not yet been widely embraced, these systems are gaining popularity as the technology develops more rapidly and the economic costs decrease as a result. Moreover, regional pollution and other environment problems caused in the past are now being treated as more serious issues, and, therefore, it is expected that new policies will more strongly regulate industries' environmental impacts in the near future. Indeed, China is yearly increasing its production with the construction of new large indoor RAS facilities (Davidson et al. 2015).

Table I.2. Species produced worldwide in RAS differentiated by continents or most important countries.

Species	US	Canada	Europe	South America	Asia
Abalone (*Haliotis asinine)*	X			X	
Adriatic sturgeon (*Acipenser naccarii)*			X		
African catfish (*Clarias gariepinus)*			X		
Arctic char (*Salvelinus alpinus)*		X			
Atlantic salmon (*Salmo salar)*	X	X	X	X	X
Channel catfish (*Ictalurus punctatus)*	X				
Coho salmon (*Oncorhynchus kisutch)*		X		X	
European eel (*Anguilla Anguilla)*			X		
Halibut (*Hippoglossus hippoglossus)*		X			
Pacific white shrimp (*Penaeus occidental)*	X				
Rainbow trout (*Oncorhynchus mykiss*)	X	X	X	X	
Seabass (*Dicentrarchus labrax)*		X			
Seabream (*Sparus aurata)*		X			
Siberian sturgeon (*Acipenser baerii)*					
Tilapia (*Oreochromis niloticus)*	X	X			
Turbot (*Scophthalmus maximus)*				X	
White sturgeon (*Acipenser transmontanus)*	X	X			

A global census of commercial RAS facilities would represent a valuable resource with respect to production volumes, market information, and a continuation of the study of the environmental impacts associated with this production.

2.6. National/international policies

Reasons, such as the lack of space for expansion and new sites (due to competition with other uses and interests), limited fresh water availability, and concerns over pollution are considered key obstacles for further expansion of conventional cage-based and flow-through (FTS) aquaculture systems (Naylor et al. 2000; Buschmann et al. 2006). Consequently, UK, Ireland, Italy, Denmark, Norway, US, Canada and Chile have promoted RAS as one of the possible solutions and opportunities to further develop aquaculture, through policies and experts (Eurostat 2011; Bellona-AquaWeb 2009) (COM 2002; COM 2009;NOAA 2011a) (De Ionno et al. 2006; Martins et al. 2010; Klinger and Naylor 2012). Moreover, environmental agencies and Conservation Funds such as the Monterey Bay aquarium´s SeaFood Watch program and The Freshwater Institute also support RAS installation and operation in federal waters of the US (NOAA 2011b; Badiola et al. 2014).

Output from the European aquaculture industry has largely stagnated over the last ten years (COM 2009; APROMAR 2010; ECF 2011). This, coupled with a continued demand for seafood, has led to Europe’s increasing reliance on imports (COM 2009). Additional reasons include already mentioned limited access to new sites and restrictions on production due to concerns over environmental impacts, hampering so the ability of European producers to compete with imports (FAO 2001). Policy priorities within the European Union place greater emphasis on food quality, safety requirements, and sustainability (i.e. economic, social and environmental) than on promoting low-cost production, as is done in other production regions. Future food production (i.e. seafood) must have sustainability as a central objective (Martins et al. 2010).

For instance, European Council 2000/60/EC of 23 October 2000 - “The Water Framework Directive”- set out a new framework for managing the quality of fresh and coastal waters that strengthens the powers of responsible agencies to manage sources of pollution and promote more “environmentally friendly” production methods. In 2002, the European Commission published a “Strategy for the Development of European Aquaculture” (COM, 2002) which provided the policy framework for revisions to European Structural Funds (i.e. European Fisheries Fund) and for research and other projects funded under the 6th and 7th RTD Framework programs (i.e. SUSTAINAQUA and CONSENSUS, respectively). However, after considering the lack of progress on the central objective of promoting aquaculture’s development, The Commission issued a new communication in 2009, giving more importance to increase competitiveness whilst retaining the focus on sustainability.

3. Aquaculture and fisheries in the Basque region

The coastal area of the Basque Country (Northern Spain) covers a surface of 20,664 km^2 and represents a population of three million people. Its economy originally based primarily on agriculture, fishing, maritime and trading activity, relies now on industry and services (Table I.3). Basque region supports one of the largest European commercial fishing industries and the production volumes have been self-sufficient to satisfy internal consumption needs. Nevertheless, fisheries and agriculture has suffered an important decline due to different factors (Zallo and Ayuso 2009) such as the reach of the maximum potential of the exploited fishing grounds and the biological extinction of populations (e.g. anchovy, hake and tuna) (ICES 2002; 2007; Sanz et al. 2008).

Table I.3 Basque region's socio-economic profile's evolution from 1930 to 2013 (Updated from Zallo 2006; Martinez-Churiaque 2007; COM 2016a).

	1930	2006	2013
Fisheries and agriculture	25%	1%	0.65%
Industry and construction	42%	38.2%	32%
Services	33%	60.8%	58.8%

Moreover, in the last years, the imports of seafood products have been higher than the produced ones and the economic values of the exports have been overtaken for the imports (A.T. 2013). Such situation has opened an opportunity for aquaculture industry's development, and local policy makers have decided to promote aquaculture planning, with the aim of creating a sustainable and complementary economic activity to the fishing and seafood sectors operating within the region (EJ-GV 2014).

Historically, aquaculture has being a minor industry in the region. The existing limiting factors for industry's development have been: (I) lack of coastal space to install farms; (II) restricting environmental rules with high administrative burden; (III) strong waves, currents and general oceanographic conditions; (IV) strong competency on food market and fish prices; (V) high investment costs and lack of interest within the local investing groups; and (VI) negative public perception of aquaculture as activity (EJ-GV 2008).

During the period 1998-2004, Basque aquaculture production experienced an average annual growth rate of 27% (whereas 4.7% and 15.8% belonged to marine and continental aquaculture, respectively) (DAPA 2008). However, between 2007 and 2013 the setback was of 81%. Currently, from the seafood consumed in the region, aquaculture represents 26 and 28% of the volume and value, respectively (Fig. I.3) (EJ-GV 2014). Moreover, aquaculture products are dominated by species such as shrimp (*Litopennaeus vannamei*), salmon (*Salmo salar*), sole (*Solea senegalensis*) and mussels (*Mitylus galloprovincialis*) (EJ-GV 2014) (Fig. I.4); hence, referring as main candidates for the aquaculture developments within the region.

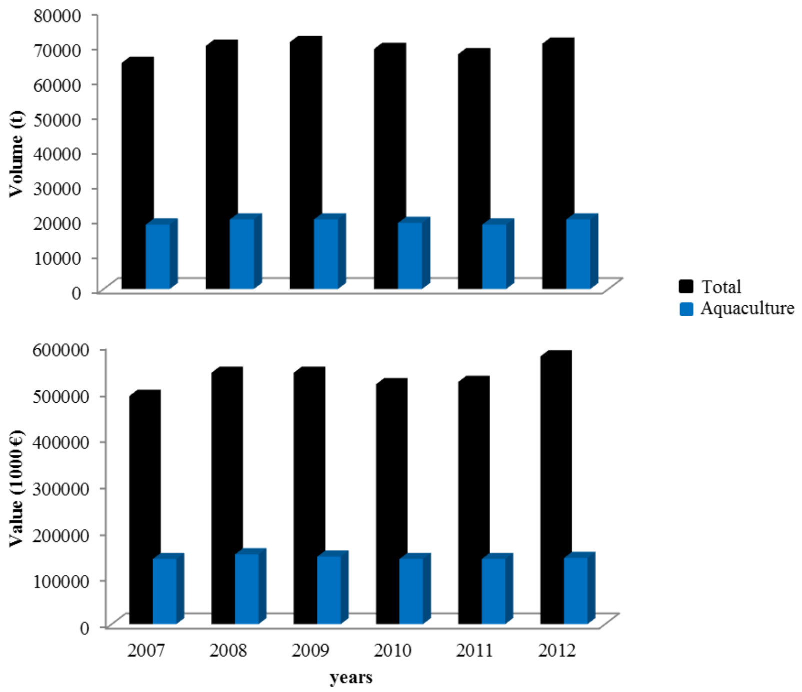


Figure I.3 Fisheries and aquaculture products' consumption evolution between 2007 and 2012 in the Basque region (EJ-GV 2014).

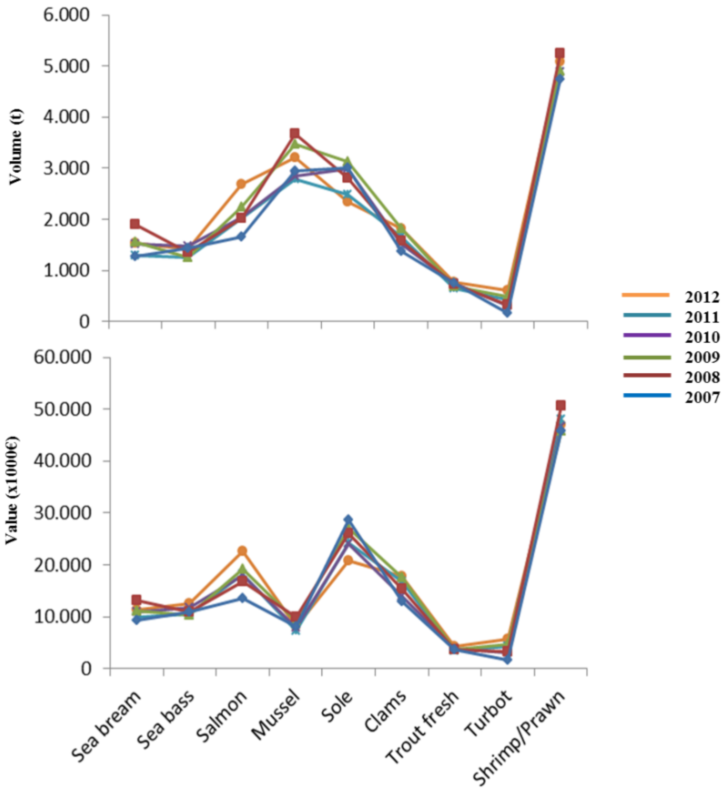


Fig. I.4. Volume (up) and value (down) of aquaculture products consumption in the Basque region (EJ-GV 2014).

In general terms, it seems clear that there is an opportunity for aquaculture as a food industry to be developed. Basque consumers are prone to eat fish products and local commercial fisheries continue declining. While seafood per capita consumption rate in Spain is of 26.4 kg/person/year, in the Basque region is of 36.1 kg/person/year (MAGRAMA 2015), which highlights the importance of fish in the Basque society. Farmed products may result less favored in terms of societal attributes comparing with wild products, but their image of being more sustainable in environmental terms could make them more attractive for the public at the medium term run (Nielsen et al. 2007). Moreover, realistic scenarios for technological change in aquaculture and institutional development in fisheries management can combine to ensure that current per capita consumption levels and sectorial economic profitability can be sustained with the right policy and investments within each corresponding sector (Merino et al. 2012). Likewise, considering that marketing and processing after the harvest are very similar between aquaculture and fishing activities, farmed products may complement the local supply outside the fishing seasons (Natale et al. 2013). Therefore, it´s expected that the inclusion of the aquaculture industry into the overall Basque food industry´s scenario would aid to add value to the whole local food chain.

Based on the Basque society’s fish consumption habits, the preference of this society to consume locally grown products, and the shown interest by local companies for particular species production, Atlantic cod (*Gadus morhua*) and salmon (*Salmo salar)* have been selected as potential species to be grown in the Basque country (EJ-GV 2014).

3.1. Atlantic cod (*Gadus morhua*)

Atlantic cod (*Gadus morhua*) (FAO 2015) represents the cornerstone of gadoid fish, being likely the most famous cold-water marine fish species, both from an economic and a socio-economic point of view (Kurlansky 1997). It is generally considered a demersal fish, although its habitat may become pelagic under certain hydrography conditions (i.e. when feeding or spawning). Its presence usually depends on prey distribution rather than on temperature. In the natural habitat it can attain ages of 20 years, sizes of 160 cm and weights of 40 kg. Its natural and characteristic skin color is brown greenish, intensively dotted. It has a big mouth with a very unique beard in the lower jaw and it is one of the most fertile marine species; each female can easily spawn between 3 and 9 million eggs in successive spawning batch (Fahay et al. 1999) (Fig. I.5).

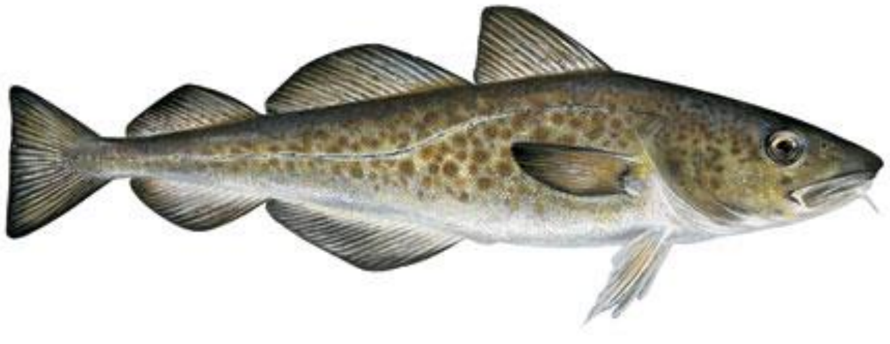
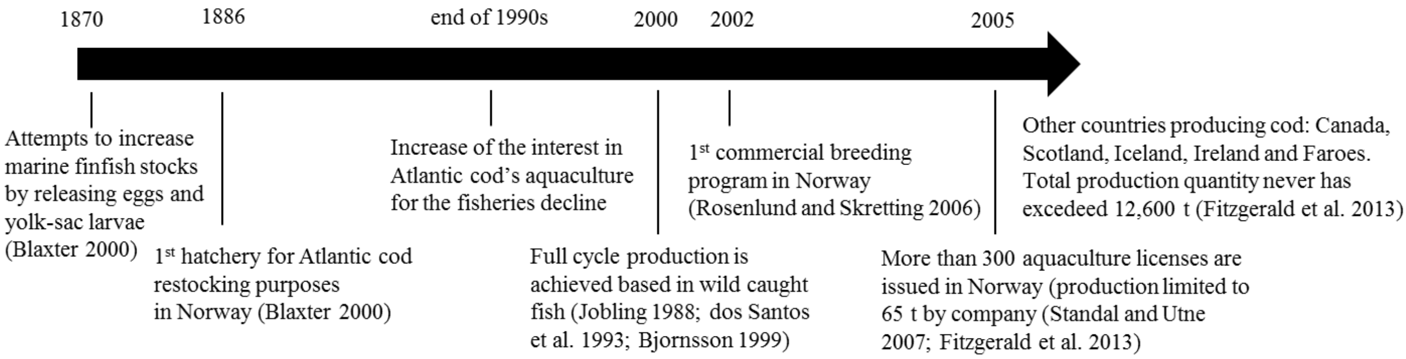


Fig. I.5. Atlantic cod (*Gadus morhua*) (FAO 2015)

Cod is distributed in several stocks taking both sides of the Atlantic: from North Cape Hatteras to the ice edge in the West Atlantic and, from the Bay of Biscay in the south to the northern part of the Barents Sea in the East Atlantic. Its annual landings declined over the years; from 2.08 million tons to 770,503 t between 1950 and 2008 (FAO 2015) due to overfishing. At present, European main cod fish stocks' situation remain relatively constant within sustainable recruitment limits; they are being sustainably exploded and both the total biomass and breeders are at maximum historic (AZTI 2014). Some cod fish stocks have remained commercially extinguished due mainly to overfishing (Esmark and Jensen 2004), and other interacting factors: ocean´s temperature variation, seals predation, and capelin (*Mallotus villosus*) prey biomass modification (Murua 2001). Currently, the allowable catch is focused in a small number of northern fishing nations (i.e. Russia, Iceland, Denmark and Norway), accounting for over two-thirds of the quota, while there are not catches serving the US and/or Canadian markets (Murua, *pers. comm.*). Thus, in terms of demand, cod markets present a huge supply opportunity and the aquaculture industry attempts to satisfy this demand.

3.1.1. The chronology of cod farming

The interest in cod farming was mainly due to salmon farming success. Figure I.6 shows the advances along the time.



Attempts to increase marine finfish stocks by releasing eggs and yolk-sac larvae (Blaxter 2000)

1st hatchery for Atlantic cod restocking purposes in Norway (Blaxter 2000)

Increase of the interest in Atlantic cod's aquaculture for the fisheries decline

Full cycle production is achieved based in wild caught fish (Jobling 1988; dos Santos et al. 1993; Bjornsson 1999)

1st commercial breeding program in Norway (Rosenlund and Skretting 2006)

More than 300 aquaculture licenses are issued in Norway (production limited to 65 t by company (Standal and Utne 2007; Fitzgerald et al. 2013)

Other countries producing cod: Canada, Scotland, Iceland, Ireland and Faroes. Total production quantity never has excedeed 12,600 t (Fitzgerald et al. 2013)

Fig. I.6. Atlantic cod's aquaculture advances along the time

Stakeholders assumed that cod farming could be the new species suited for large-scale production; more when wild stocks began to decrease and prices increased rapidly (Standal and Utne 2007). By the year 2010, it was stated that the production had reached the levels of Atlantic salmon (*Salmo salar*) production, but this was not achieved due to costs of production in comparison to the market prices (Bjornsson et al. 2010). Only Fülberth et al. (2009) have reported an attempt to date of rearing cod to marketable size using RAS. The on-growing phase is nowadays based in sea-cages production, limited by the lack of juvenile supply due to their high prices (Bjornsson and Olafsdottir 2006). In the past years, there has been a great progress to develop a steady and secure cod juvenile production and many investigations have been focused on that (Svasand et al. 2004; Bjornsson and Olafsdottir

2006; Foss et al. 2006, Imsland et al. 2007a; Remen et al. 2008; Fülberth et al. 2009; Moran and Stottrup 2011; van der Meeren et al. 2011). Final mean weights above 1 -1.5 kg have been reported using partial RAS (Lambert and Dutil 2001) and on-growing from 192 to 800 g have been also achieved (Rosenlund et al. 2004). Likewise, cod have been extensively studied for many years, albeit most of those investigations have focused on particular experimental conditions such as, stocking densities (Lambert and Dutil 2001; Bjornsson and Olafsdottir 2006; Foss et al. 2006; Bjornsson et al. 2012), thermal (Bjornsson et al. 2001; Bjornsson et al. 2007) or photoperiod treatments (Imsland et al. 2005a; Imsland et al. 2007b; Fülberth et al. 2009).

3.1.2. Cod and the Basque region

Atlantic cod´s (*Gadus morhua*) relation with Basque population began in 1670 with whales (*Eubalaena glacialis)* fisheries in the Gulf of Biscay. The intensive fishing activity in the area from other fleets (Germany, Britain and Netherlands) pressed the Basque fleet to arrive to the North Sea, Iceland, Svalvard (Norway) or Canada (Labrador and Terranova) for the fishing of cod (Garay 1985). At present, Basque cod fleet works in two fishing grounds: Terranova (Canada) and Svalbard (northern part of Norway), west and east part of North Atlantic Sea, respectively. Lately, cod´s fishing contribution to the total income of the Basque fleet has significantly decreased due to normative restrictions and quota decreases in the aforementioned grounds. Conversely, local cod fish consumption has continued increasing with time following fishery´ situation (i.e. declines and recoveries in 2008 and 2013, respectively). Therefore, yearly cod’s consumption has steadily gone increasing following the fishery’s recovery, from 2,000 to 3,500 t. Just in the Basque region, its market values represent nearly 30 million euro per annum (MAGRAMA 2014) (Fig. I.7); in fact, it represents a good candidate species for aquaculture industry.

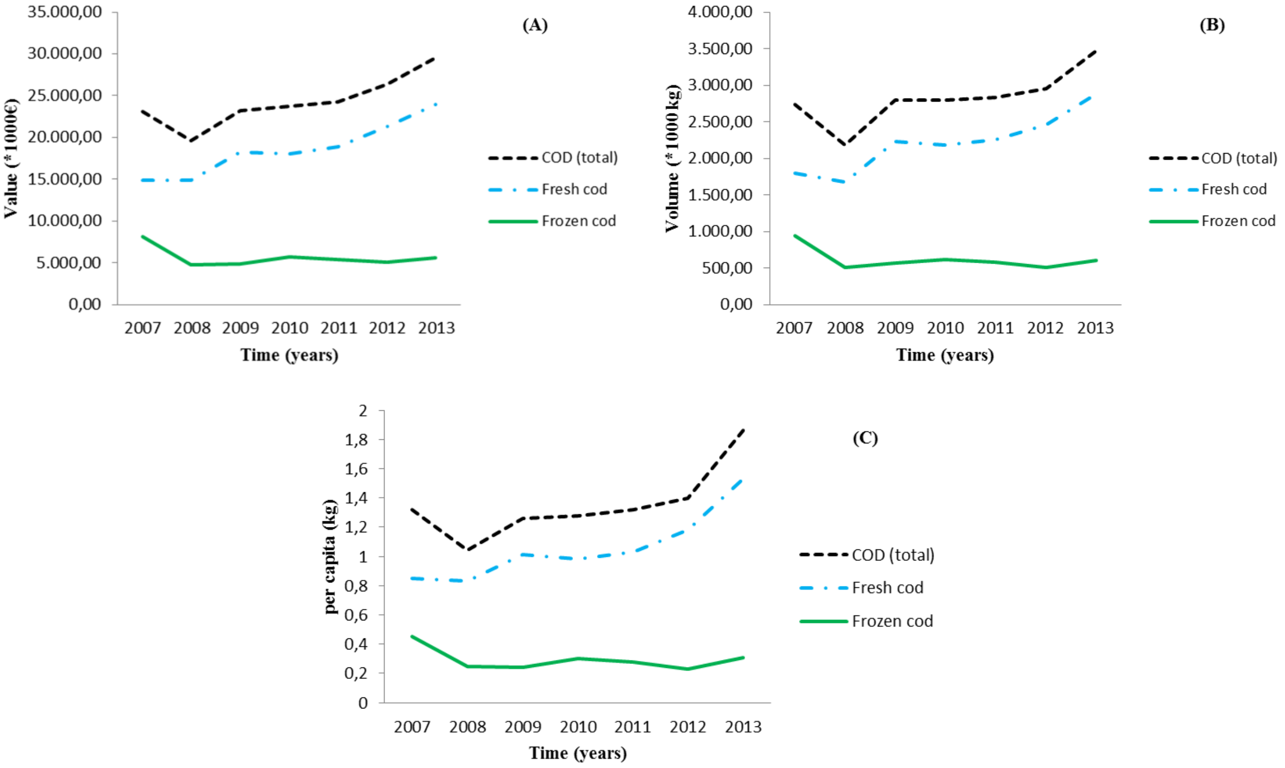


Figure I.7 Atlantic cod (*Gadus morhua*) consumed value (A) and volume (B) and per capita consumption (C), in the Basque region between 2007 and 2013 (MAGRAMA 2014).

3.2. Atlantic salmon

Atlantic salmon (*Salmo salar*) belongs to the family Salmonidae and genus *Salmo* (FAO 2016). A common characteristic is the presence of an adipose fin just in front of the tail on the dorsal (top) side. It changes its appearance during different life cycles, but in general it has an elongated body shape, which becomes deeper with age, and silvery color (Fig. I.8). In natural conditions, it can attain sizes of 150 cm and weights of 39 kg. Its life cycle is complex: it is anadramous (i.e. spent part of their lives in fresh water and the other part in salt water) and has evolved a finely tuned “chemotactic” system allowing it to return for spawning (5th-6th year of age) to the very stream in which they were hatched. Juveniles take mainly aquatic larvae of insects and in the sea they are feed with crustaceans and small fishes (i.e. herring, sprat, sand-eels, capelin and small gadids). At the sea it has small scales, with silvery flanks and a white belly, while after migrating to fresh water it color changes, becoming greenish or brown and red or orange mottled. Additionally, males develop an exaggeratedly hooked lower jaw, and a staggered line of teeth.

Fig. I.8. Atlantic salmon (*Salmo salar*) (COM 2015)

Atlantic salmon (*Salmo salar*) is North Atlantic's native species consisting of three populations: North American, European, and Baltic. Their geographical distribution goes from the Atlantic coasts of Europe, from Barents Sea, northern Norway and Baltic southward to northern Portugal, also around Iceland and southern Greenland; not in Mediterranean. Elsewhere, coasts of Canada and North America (FAO 2016).

3.2.1. The chronology of salmon farming

The culture of salmonids (particularly Atlantic salmon-*Salmo salar*) is one of the most important examples of commercially successful intensive aquaculture productions in the world. Its potential for farming is excellent since it: (I) is relatively easy to handle; (II) grows well under culture conditions; (III) has a relatively high commercial value; and (IV) adapts well to farming conditions outside its native range. Salmon farming is a demonstration of what can be achieved through conscious investment, innovative research, technological advances and creative marketing strategies. At the same time, it has served to illustrate the dangers of rapid development and depressed prices that result when market capacity to absorb increasing supplies is exceeded.

Salmon farming started on an experimental level in the 60s in Norway (Laird 1996; Torrissen et al. 2011) at the time that wild captures began to decline (Fig. I.9).

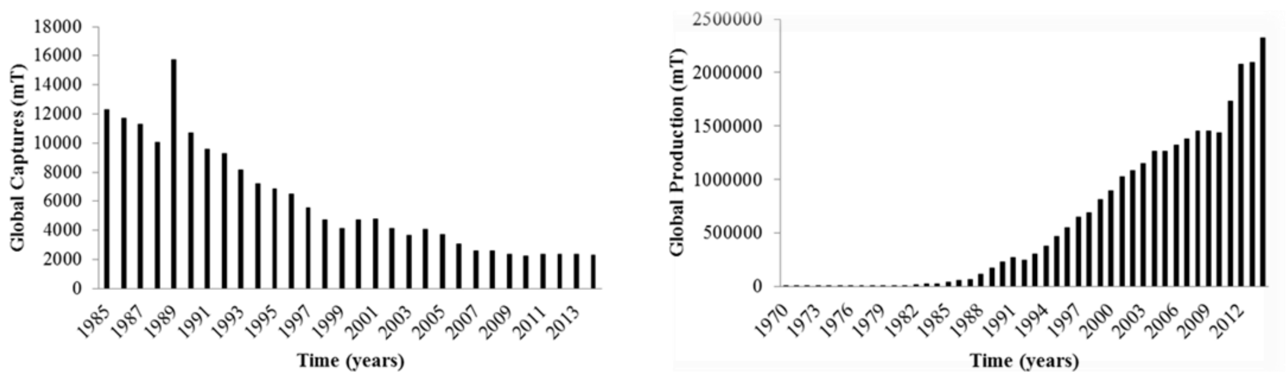


Fig. I.9. Atlantic salmon (*Salmo salar*) capture and global production (million t).

The chronology of salmon farming is presented in Fig. I.10. Currently, the production of salmon smolts for recruitment in sea cages takes place mainly in Europe (i.e. Norway, Scotland, the Faeroe Islands, Iceland and Ireland) although over the past decade, the United States have also increased the interest in establishing and using land-based, closed-containment systems (e.g. RAS) for salmonid culture, particularly Atlantic salmon (Burr et al. 2012).

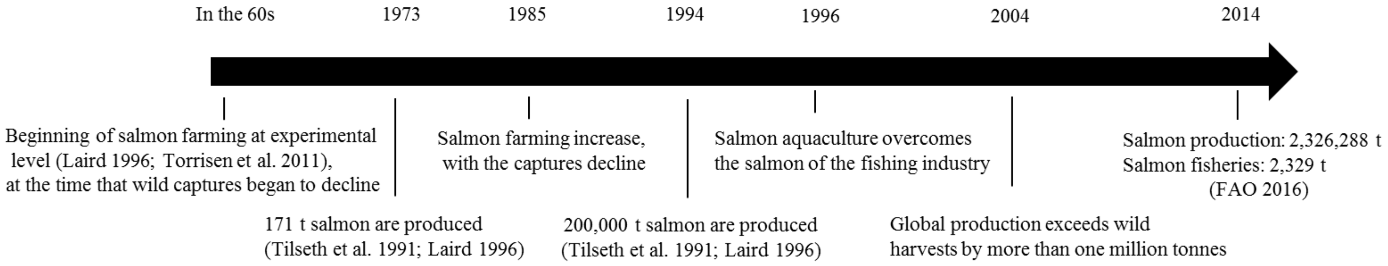


Fig. I.10. The chronology of salmon farming.

In Europe, the dominate producer is Norway accounting for some three quarters of the total annual production (Bergheim et al. 2009), with an annual output close to 250 million smolts. In Scotland, salmon production increased from 14 to 163,000 t between 1971 and 2013 and it is currently the largest farmed salmon producing country in the European Union. Ireland had been an important producer for the European and US market since 1980. Nevertheless, the industry did not develop and the production dropped from 25,000 to 10,000 t between 1990 and 2015 (Warrer-Hansen 2015). Apart from European countries, Chile had an exponential growth, becoming the second largest producer in less than 20 years; by the end of 2004 the production of farmed salmonids reached 550,000 t (Buschmann et al. 2006). Growth occurred with scarce regulation (Asche et al. 2009), which led the industry to suffer two of the most dramatic collapses in the salmon industry: the occurrence of the Infectious Salmon Anemia in 2007, causing the caused production's decrease from 400,000 to 100,000 t between 2005 and 2010, respectively; and, the deadly algal bloom in 2016, mainly due to "El Niño" which affected 20% of the production.

Salmons, when juvenile, are changed from land-based hatcheries into sea-cages. This hatchery production has been based in flow-through systems to date. Nevertheless, available data suggest that it is shifting towards RAS technology, due mainly to: (I) poor performance during on-growing in the seawater cages (Bergheim et al. 2009); (II) lack of available water supply (Joensen 2008; Kristensen et al. 2009); (III) large season variation in water temperature and low inlet water quality (including aluminum concentrations) (Kristensen et al. 2009); and (IV) increased quality (growth and survival after sea transfer) of RAS-cultured smolts (Terjesen et al. 2008; Martins et al. 2010). An example is the production of smolts in the Faeroe Islands, where a complete shifting from flow-through into RAS took place after 2000 (Bergheim et al. 2008; 2009). Thus, all seven active smolt farms on are RAS and an increase of smolt size from 50 to 70 g from flow-through farms to 140–170 g in RAS was reported (Joensen 2008). In Norway, the productivity at hatcheries-smolt farms has increased as well. In 1985, a typical farm was producing 100,000-300,000 smolts/year of 30-50 g at delivery; in 2000 this production increased to 500,000-2,000,000 smolts/year of 70-120 g (Bergheim and Brinker 2003).

Globally, farmed salmon industry has grown substantially in the past 40 years and today; approximately 60% of world's salmon production is farmed. In fact, Atlantic salmon is the highest value species in Europe, accounting for 20.9% of the total value (COM 2016). Moreover, salmon consumption worldwide is currently three times higher than it was in 1980, which have made the salmon farming industry to be the fastest growing food production system in the world accounting for 70 % (i.e. 2.4 million tons) of the market.

3.2.2. Salmon and the Basque region

Atlantic salmon has migrated through Basque rivers for decades but it has never been farmed in the region. As shown in Fig. 5, its consumed volume increased from 1,500 to 2,800 t, while its value was doubled from 11,000 to 23,000 k€ between 2007 and 2012. In fact, salmon consumption has been the fastest growing one among the most popular species for the Basque consumers.

One of the reasons for such increase is the boost of the Japanese food in general, sushi in particular, worldwide. In the last 5 years, there has been a boost of Japanese restaurants and shops both in Spain and in the region. One of the main characteristics that differentiate such restaurants/shops is the need of very good quality and fresh products (i.e. they are eaten raw) available anytime. As result of a survey made by the author among 10 sushi restaurants and/or shops in the area, local salmon could be a good marketing strategy for product (i.e. sushi) selling. Currently, employed salmon is farmed (due to: lower prices than wild caught salmon; product available year-round; quality and size) from both Norway (in 7 cases) and Scotland (in 3 cases) and it is purchased once week. Offered product's price is pointed out as the only dubious factor while the possibility of having fresh salmon any time along the week creates interest among the interviewees.

Salmon presents an interesting framework as species to be farmed in the region.

4. Feasibility studies for aquaculture industry's development

In 2007 and 2014, the Basque government designed a strategic plan for aquaculture's development within the region (EJ-GV 2008; EJ-GV 2014). RAS were proposed as a strategic technology for sectorial development. As abovementioned, there are various aquaculture groups interested in commercially producing aquatic species in the Basque region. Nevertheless, to assess the feasibility of a newly designed economic activity within a particular location such as aquaculture in the Basque region, different aspects should be primarily studied. A feasibility study is made to determine the potential of a given activity. It is generally conducted during the early stages of a project analyzing different scenarios and factors that can influence both the business and its subsequent sustainability (economic, environmental and societal) (Amanor-Boadu 2009). Additionally, the environmental concern over the aquaculture industry makes necessary to study each activity in detail, reporting key aspects for success towards a sustainable production: (I) technical and technological viability of RAS and species reared; (II) economic viability of the systems; (III) social acceptance of the produced fish; and, (IV) environmental friendliness of the systems with low impacts (Fig. I.11). In Fig. I.11 it is represented the "positive relationship" between: the competitions for the existing natural resources, socioeconomics needs, primary production opportunities, development of engineering, and global economy. Herein, a sustainable production is achieved when the three pillars of sustainability (i.e. society, environment and economy) converge.

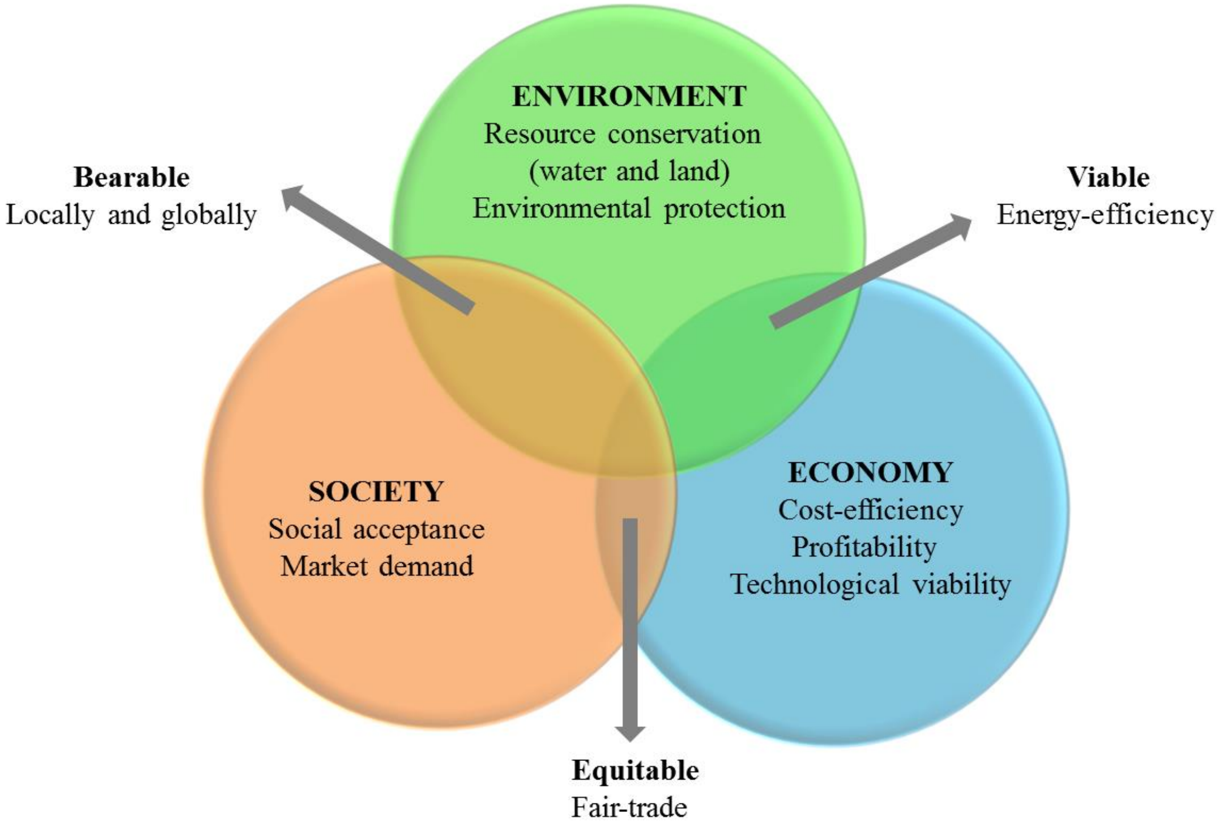


Figure I.11 Three conceptual elements of sustainable development required for a sustainable animal farming production.

The operation of RAS requires education, expertise and dedication (Dunning et al. 1998). Prospective operators of RAS need to know about the required water treatment processes, the component of each process and the technology behind each component. Many commercial RAS have failed because of component failure due to poor design and inferior management (Masser et al. 1999; Timmons and Ebeling 2010; Badiola et al. 2012). A correct knowledge on system´s design, specification of technical components, and general operations is a minimum prerequisite for the success of a RAS farm (Badiola et al. 2012). Capital investment for the setup of a RAS farm is normally much higher than that of a conventional production system, due to the requirement for additional equipment to treat water for reuse. The water treatment process can increase operation costs and secure the production volumes but every failure at that level also result in huge economics losses (Summerfelt et al. 2001). Therefore, the economic aspects has to be taken into account before embarking on any other consideration (Bijo 2007) .

Hypothesis and objectives

1. The hypothesis

The following hypothesis is posed as a basis of this thesis:

“The farming of cod and salmon as cold water marine species might be feasible using Recirculating Aquaculture Systems (RAS) in the Basque coastal area (Northern Spain) in a technically, environmentally, economic and socially sustainable way using local seawater’s temperature profile”.

2. The objectives

In order to prove the aforementioned hypothesis, the present work attempts to address the following general objective:

To evaluate if the farming of cod and salmon as cold marine species is feasible using Recirculating Aquaculture Systems (RAS) in the Basque coastal area (Northern Spain) in a technically, environmentally, economic and socially sustainable way using local seawater’s temperature profile.

The general objective has been subdivided in a series of specific objectives as follows:

Technical

1. to analyze the **influence of local seawater temperature** conditions on cod and salmon growth and mortality.
2. to define **critical points on management operations** of Recirculating Aquaculture Systems.

Social

3. to determine grown-out cod and salmon individuals **nutritional profile**.
4. to determine **social acceptance** of grown-out cod and salmon product reared in the Basque Country, with reference to wild caught individuals and/or other farmed fish approaches.

Economic

5. to describe the structure of the **economic costs and cost-benefit scenarios** of setting up a Recirculating Aquaculture System to rear the target species of the present study.

Environmental

6. to **improve the environmental assessment** methodology and thus, the economics of RAS.
7. to **assess the key energy challenges** of RAS.
8. to evaluate the **environmental impacts** (cradle-to-grave) created while rearing cod in RAS in the Basque region.

Overall

9. to **propose a more efficient** RAS to decrease environmental impacts and economic costs in order to produce fish in a more sustainable way.

4. Working plan

In order to achieve the mentioned objectives a structured working plan was outlined. The experiments relating to cod and salmon farming followed the same working protocol as explained in the Experimental Design section, which resulted in the species growth and survival, final product's organoleptic profile, and its social acceptance and systems' overall performance reflected in Contributions 1 and 2.

RAS are high energy demanders due to the continued pumping of the incoming water and its treatment. Nevertheless, there is a lack of information around the energy usage in the industry. Thus, Contribution 3 resulted in a review that compiled published data and information (i.e. where in the system the energy is needed, energy sources used in the industry) from articles and books, a questionnaire comprehending industry's point of view about the energy and its importance, and a guideline to design a more energy efficient RAS.

Life Cycle Assessment (LCA) methodology studies the environmental impacts created during a given process and they help, among others things, to identify a processes' environmental impact hotspots. On the other hand, energy audits identify the energy flows of a system, and take into account diverse parameters of the production such as in aquaculture temperature, reared species, and help to draft an energy use diagram which results in proposing energy saving solutions. Thus, Contribution 4 resulted in the combination of the two mentioned methodologies (i.e. LCA and energy audits), which would help in the identification of where, within the pilot-scale RAS units, was the energy used (e.g. which equipment consumes the most), and identify areas where energy can be saved in RAS to improve both the environmental and economic performance of the systems.

Many RAS have been built over the years but many systems and/or companies have failed due to different reasons, such as economic issues and fish marketing problems. Furthermore, RAS are high technology systems that requite a combination of knowledge (i.e. fish biology and husbandry, engineering of the systems, water quality), which has to be applied all together. A worldwide questionnaire (including researchers, consultants and industry managers) was performed in order to understand the main issues in the management of RAS and future challenges, resulting in Contribution 5.

For each specific objective mentioned in Section 2, several challenges were addressed, which are included in the following Table O.1:

Table O.1 Challenges addressed per each of the thesis objectives in each of the contributions

Contribution Nº	**Feasibility dimensions**	**Specific objectives (related to pages 1 and 2)**	**Targeted species**	**Related challenges**
1	Technical Economic Social	1, 3, 4, 5	Cod	• Individuals adequate growth performance • Look for operational and investment costs • Profitable and cost-effective production • Affordable product prices • Consumers acceptability of the locally grown products
2	Technical Social	1, 3, 4	Salmon	• Individuals adequate growth performance • Affordable product prices • Consumers acceptability of the locally grown products
3	Technical Environmental Economic	6, 7, 9	No specific species targeted	• Management issues and consequences • Energy usage of the systems and possible improvements • Types of energy sources • Further investigations
4	Technical Environmental	6, 8	Cod (although applicable to any other species)	• Reducing the environmental impacts of RAS • Looking for sustainable alternatives
5	Technical Environmental	2, 9	No specific species targeted	• Management issues • Critical points in RAS from different perspectives • Future challenges

4. Contributions

The results from different works carried out during the development of this thesis have led to a series of presentations in international congresses and publications in different peer-reviewed journals. Some of them represent the main core of the present thesis; others have served to give context to the followed lines of work. A number of them have already been published and others are accepted for publication. The most relevant ones are listed below according to the Contributions of this thesis. The others such as posters, technical publications and diverse congress participations are listed in the Annex 1 at the end of the thesis.

Contribution I) **Badiola, M.,** Albaum, B., Mendiola, D. (2016). Land based on-growing of Atlantic cod (*Gadus morhua*) using Recirculating Aquaculture System (RAS); a case study from the Basque region (northern Spain). *Aquaculture* 468, 428-441.

- **Badiola, M.,** Cabezas, O., Curtin, R., García, M., Gartzia, I., and Mendiola, D (2015). On-growing of Atlantic cod (*Gadus morhua*) to marketable size in a land-based system – a feasibility study from the Basque Country (Northern Spain). *RIMER 2015*. Invited speaker. Donostia-San Sebastian (Spain). February 8th.
- **Badiola, M.,** Cabezas, O., Curtin, R., García, M., Gartzia, I., and Mendiola, D. "Estudios de viabilidad sobre el crecimiento y produccion del bacalao (G*adus morhua*) hasta talla comercial". *Dia de la Acuicultura*. Invited speaker. PIE-Plentzia (Spain). November 28th 2014.
- **Badiola, M.,** Cabezas, O., Curtin, R., García, M., Gartzia, I., and Mendiola, D. "Land based on-growing of Atlantic cod (*Gadus morhua*) to marketable size – a feasibility study from the Basque Country (Northern Spain)". *Aquaculture Europe 2014*. Donostia-San Sebatián (Spain) October 14-17.

Contribution II) **Badiola, M.,** Gartzia, I., Basurko, O.C., Mendiola, D. (2017). Land-based growth and sensory evaluation of Atlantic salmon (*Salmo salar*) in Northern Spain: looking consumers acceptance. *Aquaculture Research* (accepted, DOI expected for Sept. 2017)

- **Badiola, M.,** Basurko, O., Gartzia, I., Mendiola, D. (2015). Marketable size salmon (*Salmo salar*) production in the Basque region: a feasibility case study from a Recirculating Aquaculture System (RAS). *Aquaculture Europe 2015*, Rotterdam (The Netherlands). Conference paper. October 20-23.

Contribution III) Badiola, M., Hundley, P., Basurko, O.C., Piedrahita, R, Mendiola, D. (2017). Energy use in Recirculating Aquaculture Systems (RASs): a review. *Environmental Science & Technology* (under revision).

Contribution IV) Badiola, M., Basurko, O.C., Gabiña, G., Mendiola, D. (2017). Integration of energy audits in Life Cycle Assessment (LCA) methodology to improve the environmental performance assessment of Recirculating Aquaculture Systems (RAS). *Journal of Cleaner Production* (under revision).

- **Badiola, M.,** Basurko, O., Gabiña, G., Mendiola, D. (2015). A combination of environmental assessment methods and key economic factors to improve RAS's sustainability. *Aquaculture Europe 2015*, Rotterdam (The Netherlands). Invited speaker by Biomar. Conference paper. October 20-23

- **Badiola, M.,** Cabezas, O., Gabiña, G., Mendiola, D. (2015). Combination of Life Cycle Assessment and energy audit to reduce the environmental impacts of rearing cod in a pilot scale Recirculating Aquaculture System. *LCM 2015*. Bourdeax (France). Poster. August 31st August- September 3rd.

Contriution V) Badiola, M., Mendiola, D., Bostock, J. (2012). Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges. *Aquacultural Engineering.* 51, 26-35.

- **Badiola, M.** (2013). "Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges". *Workshop - Finfish nutrition and aquaculture technology at the crossroads*. Invited speaker. Bremenhaven (Germany). 18th February.

Materials and Methods

Common working procedures followed in both cod and salmon experiments are presented in the following section in order to avoid repetition within the results section. Both experiments followed same water and biological sampling schemes, final product's acceptability studies and same RAS facilities (e.g. location, working units).

1. Location

The experiments were carried out in pilot-scale RAS units (IRTAmar®) that are commonly used in R&D projects at a private fish company owned by AZTI, in the coastal area of the Basque region (Getaria; Northern Spain) (Fig. M.1). In this area, annual seawater temperature ranges between 7 and 21ºC (Goikoetxea et al. 2009). The Cod experiment lasted from April 2011 to June 2012 and the salmon experiment from April 2013 to September 2014. The annual seawater's temperature that occurred during both experiments is presented in Fig.M.2.

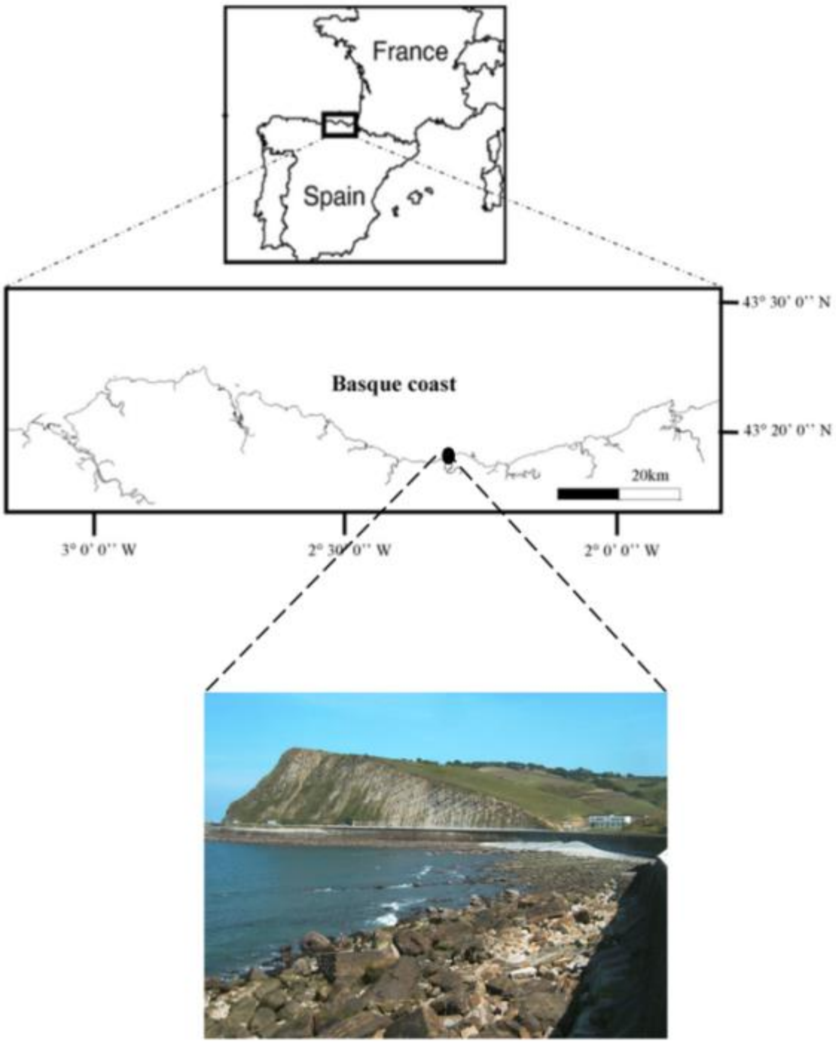


Fig. M.1. Location of the private fish farm along the Basque coast.

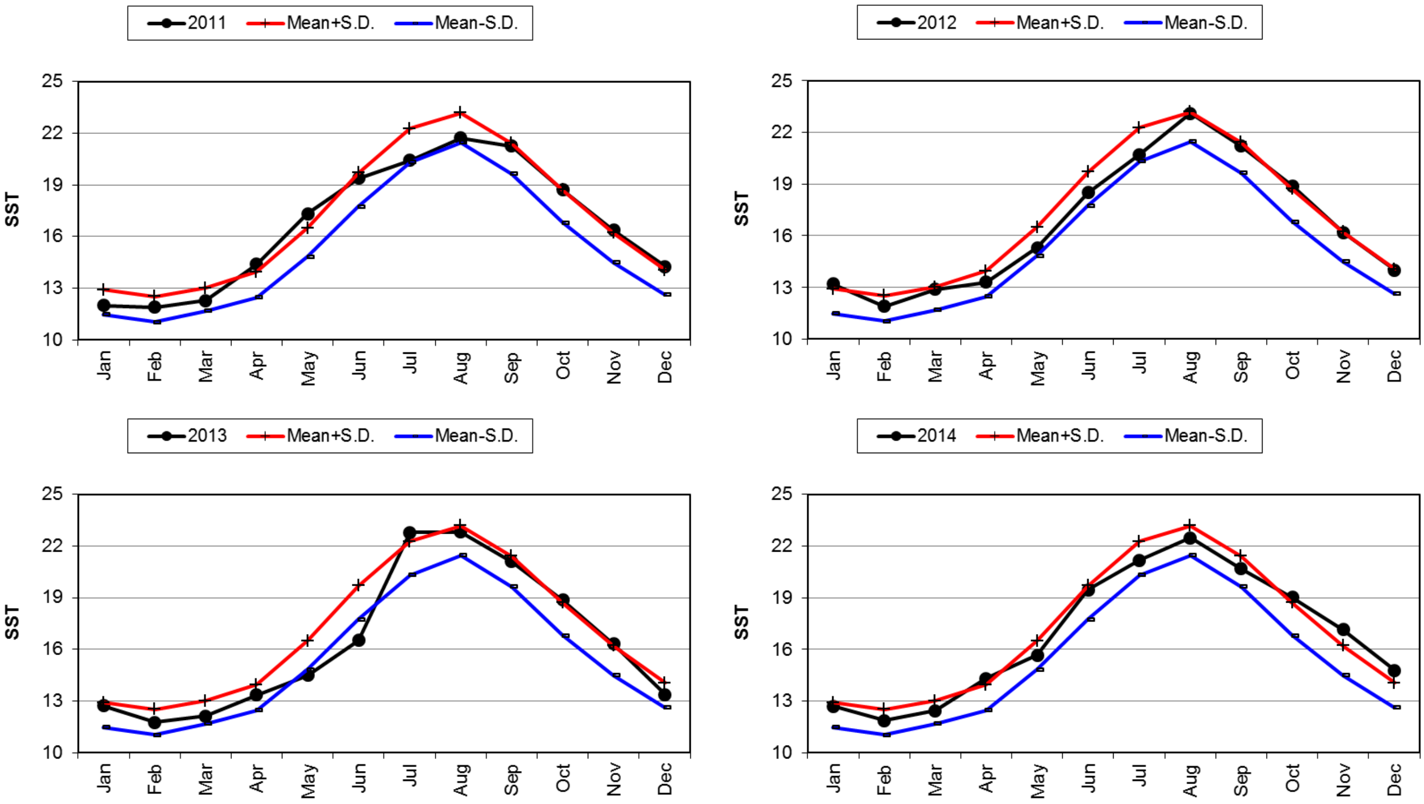


Fig. M.2. Seawater Surface Temperature (SST) in ºC during experimental years 2011, 2012, 2013 and 2014 in the local area.

2. Pilot-scale RAS description

In both experiments (hereafter cod experiment as first, and salmon experiment as second) 2 pilot-scale RAS units were used in parallel, consisting of 3 blue tanks (3 m diameter and 7 m^3 volume each) with a total working volume of 24 m^3, and a water treatment loop (Fig. M.3).

Incoming water was pumped in directly from the sea by 2 pumps through a sump located 470 m away from the farm itself. The water was pre-treated for solids removal and ozonizated before going through the rearing tanks.

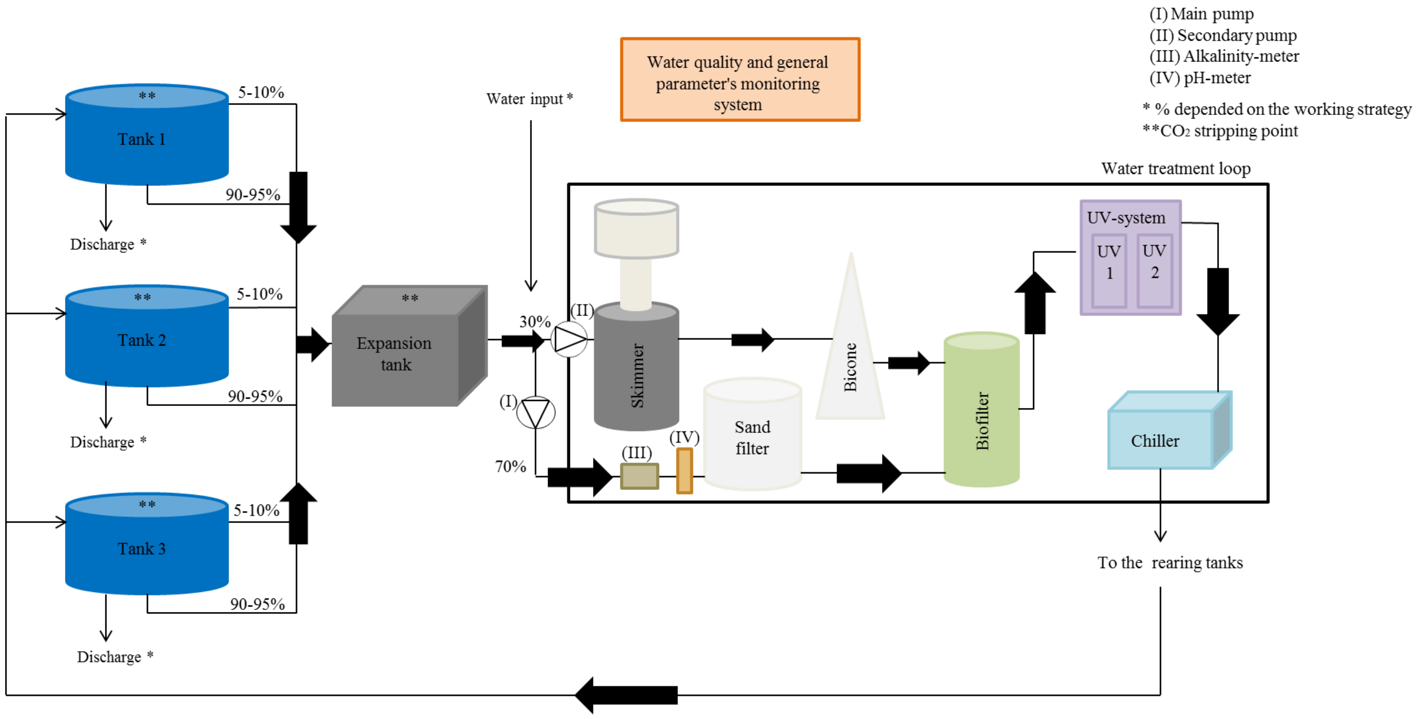


Fig. M.3. General overview of the pilot-scale RAS unit.

Tanks were of the dual-drain design, meaning that the majority of water (90-95%) was collected at the bottom of the tanks and the remainder (5-10%) from the top side; obtaining two flows per tank. Then, both flows entered the treatment loop. This particular design was aimed to avoid water overflowing, and allowed for the best possible hydraulics within the tanks to facilitate waste collection. The mentioned flows per each tank (i.e. top and bottom flows of the 3 tanks) ended in an expansion tank of 2 m^3 in volume (designed to capture uneaten feed and feces, which were removed every three days, and to function as a sump). From here, 30% of the water was directed to a skimmer and after through a bicone; the remainder 70% ran through a sand filter. Both flows ended in a pressurized biological filter. From here, water ran through a UV system and it was directed, when necessary, to a temperature exchange unit (i.e. chiller) before re-entering the rearing tanks. Within the system two CO_2 stripping points were installed: (I) in the expansion tank, and (II) just before the entrance into the rearing tanks. Water quality (i.e. temperature, salinity, pH) and system´s general parameters (including consumed kWh) were monitorized in a data cumulative meter. The data acquisition frequency was set to be every 10 seconds, resulting in more than 6 and 7 million data in cod and salmon experiments, respectively. In case of emergency, an alarm was activated and connected to a phone 24 hours. Moreover, equipment turning on/off for cleaning and maintenance purposes was done from a control panel. Finally, each unit had water quality measurement gauges (i.e. pH-meter, alkalinity-meter, and dissolved oxygen-meter), and performance sensors (i.e. water flow sensor). Described equipment, gauges and sensor's main specifications are specified in Table M.1.

Pilot-scale RAS units maintenance was completed daily, which included checking water quality parameters and overall performance. Moreover, equipment cleaning and sensors calibration were made in a scheduled order: the skimmer cleaned every week, the sand-filter backwashed every two days, pH-meter and alkalinity-meter calibrated every 2-3 weeks, and oxygen and water flow sensors calibrated monthly.

Table M.1. Equipment specifications for the pilot-scale RAS unit.

Equipment	Purpose	Quantity (per unit)	Model	Filtering surface	Manufacturer	Country
			Pilot-scale RAS unit equipment			
UV system	Water sterilization	2	UV Active 65W	-	FIAP	Germany
Biofilter[1]	Biological filtration	1	950-75	0.70 m^2	Astrapool	Spain
Sand-filter[2]	Solids filtration	1	Cantabric 900	0.64 m^2	Astrapool	Spain
Skimmer	Foam fractionation	1	Neptun 10.000	-	Ratz Aqua & Polymer Technik	Germany
Bicone[3]	Oxygenation	1	-	-	Calderería Plástica URSA	Spain
Principal pump	Water pumping	1	Columbia 3CV III	-	Astrapool	Spain
Secondary pump	Water pumping	1	Sena 1.25CV III	-	Astrapool	Spain
Chiller	Temperature exchange unit	1	Optima-15 II	-	Astrapool	Spain
			Water quality parameters gauges/sensors			
pH-meter	pH measurement	1	DulcoTest-PHER 112 SE		ProMinent	Germany
Alkalinity- meter	Alkalinity measurement	1	Optima PRO Type B		Astrapool	Spain
O_2-meter	Dissolved oxygen measurement	1/tank	420		OxyGuard	Denmark
Water flow sensor	Continuous flow measurement	1	8020		Burkert	Germany

[1] designed for working pressures of 2.5 bar.

[2] granule diameters oscillated between 0.4-0.8 mm

[3] designed by a local manufacturer on-demand

Experimental design

Cod and salmon are both cold water species. In the Basque coast, seawater's temperature fluctuates around 10-12ºC along the year, (as shown in Fig. M.2), reaching above 20ºC during summer periods (i.e. less favorable local thermal period for the targeted species). Different growth studies of several fish species have shown a decrease in the optimal temperature with weight (Brett 1979; Cuenco et al. 1985; Pedersen and Jobling 1989; Imsland et al. 1996; Bjornsson et al. 2001). Moreover, it has been proven that fish should be reared at stepwise temperature regimes, mimicking natural oceanic conditions and fluctuation. This leads to compensatory and higher growth rates (Hanson 1996; Björnsson and Steinarsson 2002). Thus, an early environmental manipulation (i.e. Tº) could create benefits in commercial aquaculture (e.g. faster growth in a given timeframe) (Luczkovich and Stellwag 1993; Imsland et al. 2005b); although there are some authors discouraging this practice, and propose further investigations to prove such advantages.

In such context, both experiments' chronologies were set in line with the natural seawater's temperature regime to accomplish a gradual biological coupling of individuals to the thermal conditions of the region. At the same time, two thermal regimes were established in each experiment in order to compare the possible benefits of the mentioned compensatory growth with a non-manipulated growth. These thermal regimes were named as "natural module" (N) (module that used water without artificial temperature control) and "control module" (C) (module with artificially controlled water).

Maximum biological temperature thresholds for the Control modules were established in 21ºC for cod (Bjornsson et al. 2012) and 19ºC for salmon (Handeland et al. 2008). Thus, the strategy used, to take advantage of the seasonal suitable thermal conditions for both species, consisted of the seawater's temperature management being achieved through two different working methodologies: chilling the water with the usage of a chiller, and/or opening/closing the incoming water's flow. Experimental conditions designed specifically for each of the species are explained in the Results section: *Contribution 1* for cod, and *Contribution 2* for salmon.

3. Water quality sampling

The range of main physical and biochemical water quality parameters (i.e. water temperature, salinity, dissolved oxygen, ammonia, nitrite, nitrate, formaldehyde, bromide, chlorite, pH, turbidity, and gas saturation) was analyzed on a weekly basis (every Wednesday) in the laboratory. In each unit, water samples were collected at the inner part of the tanks (located at the end of the water treatment loop) and the expansion tank.

Temperature, salinity, oxygen and pH were analyzed using a multiparametric meter (*YSI 556 Water Quality Meter, US*). This meter measured temperature by a precision thermistor, calculated salinity from conductivity and temperature, calculated the dissolved oxygen through a steady state polarographic and measured pH by a glass combination electrode.

CO_2 was measured by a carbon dioxide analyzer (*OxyGuard CO_2 analyzer*). Turbidity was measured through a portable turbidimeter (*HACH 2100Q, US*) using the Determination of Turbidity by Nephelometry of the US EPA water analytical method (EPA 2016). Total gas pressure was measured by a total gas pressure meter providing measurements for Total Gas Pressure (mmHg or % saturation), delta P (TGP-BP), as well as barometric pressure (BP) (mmHg), and temperature (degrees ºC) (*PT4 Tracker Total Gas Pressure (TGP) Meter, Eagar Inc., USA*). Finally, ammonia, nitrite, nitrate, formaldehyde, bromide and chlorite concentrations were measured using a spectrophotometer (*HACH DR5000, US*) by the Nessler, Diazotization, Cadmium Reduction, MBTH (adapted from Matthews and Howell 1981), DPD and Mercuric Thiocyanate methods, respectively. The mentioned methods were integrated in the spectrophotometer conforming to US-EPA approved methods (EPA 2016). Throughout all of these analyses, the test water sample was compared with a blank reagent.

The equipment used for water quality parameter measurements are detailed in Table M.2. Moreover, for each of the mentioned parameters the threshold limits were specified based on a literature (Table M.3). Some of the limits are not specific for species under this study (i.e. cod and salmon), as this was the first attempt of on-growing cod in land-based RAS, and the first attempt of on-growing salmon in the northern Spain.

Table M.2 Details of pilot-scale RAS unit conforming devices and equipment used for water quality analysis.

Measured water quality parameter	Equipment used	Model	Manufacturer	Country
Temperature, salinity, dissolved oxygen, pH	Multiparametric meter	556	YSI, a xylem brand	US
CO_2	CO_2 analyzer	CO_2 portable	Oxyguard	Denmark
Turbidity	Portable turbidimeter	2100Q	HACH	US
Gas pressure	Total gas pressure meter	PT4 Tracker	Eagar Inc.	US
Ammonia	Spectrophotometer	DR5000	HACH	US
Nitrite	Spectrophotometer	DR5000	HACH	US
Nitrate	Spectrophotometer	DR5000	HACH	US
Formaldehyde	Spectrophotometer	DR5000	HACH	US
Bromine	Spectrophotometer	DR5000	HACH	US
Chlorite	Spectrophotometer	DR5000	HACH	US

*constructed according to the pilot-scale unit´s flow-rates

Table M.3. Water quality parameters thersholds.

Water quality parameter	Threshold	Reference
Dissolved oxygen (mg/l)	5	Brett 1979
Ammonia (mg/l)	4.5	Foss et al. 2004
Nitrite (mg/l)	1.4	Pillay and Kutty 2005
Nitrate (mg/l)	-	-
pH	6.5-7.5	Coll 1986; Timmons and Ebeling 2010
Chlorite (mg/l)	0.0005	CCME 2007
Bromide (mg/l)	0.0005	CCME 2007
Gas saturation (%)	104	Gunnarsli et al. 2008
CO_2 (mg/l)	15-20	Moran and Stottrup 2011; Moran et al. 2012
Formaldehyde (mg/l)	40	Fredricks 2015

4. Biological sampling

Biological sampling was undertaken to study the performance and welfare of the on-growing individuals by measuring their weight and length, and both condition and hepatosomatic indexes were noted.

To do so, 20-30 individuals were randomly sampled from each pilot-scale RAS unit every 2.5 months, approximately. In all cases, feeding was stopped 24 hours before the sampling in order to have an empty stomach, and, thus, a real body weight. Immediately after being caught, fish individuals were anesthetized with 30 mg/l clove oil in seawater. Once in the laboratory, body length (mm), body weight (g) and liver weight (g) were calculated to the nearest 0.1 cm and 0.1 g respectively, using an ictiometer and precision microbalance (Mettler Toledo). Fig. M.4 shows some of the sampled individuals.

Fig. M.4. Cod and salmon individuals for the biological samplings: (A), (B) and, (C) different size cod individuals sampled in different sampling days; (D) and (E) gutted fish and its stomach; (F), (G), (H) and (I) different size salmon individuals sampled in different sampling days.

Statistical methods employed to estimate fish growth:

Instantaneous rate of growth (G) and the relative rate of growth (K) were estimated as follows (Ricker 1975),

Instantaneous body weight growth: $W_t = W_0 * e^{Gt}$ (eq.1)

Instantaneous body length growth: $L_t = L_0 * e^{Gt}$ (eq. 2)

Relative rate of growth: $K = e^G - 1$ (eq. 3)

where W_0 and L_0 are the initial body weight (g) and length (mm), respectively and W_t *(g)* and L_t (mm) are weight and length at time t (days), respectively. The daily specific growth rate was defined by K x100 (%). Data per each pilot-scale RAS unit were fitted, by linear regression, to test the suitability of the exponential model (Laurence 1976).

The condition factor (CF) (i.e. the relation of body weight and length influenced by fish age, sex, season, stage of maturation, fullness of gut, type of food consumed, amount of fat reserve and degree of muscular development) was defined as:

Condition factor: $CF= 100 * W * L_T^{-3}$ (eq. 4)

where W is the weight of the fish and L_T the corresponding total length.

The hepatosomatic index (HIS) (i.e. indicator of the energy reserve status of the individuals) was calculated as:

Hepatosomatic index: $HIS = L_W/T_{W*}100$ (eq. 5)

where L_W is the liver´s weight and T_W the total body weight.

Mortality (M) as a % per month was calculated in the following way:

Mortality: $M = 3000 (\ln N_0 - \ln N_1)/d$

where N_0 was the initial number of fish, N_1 the final number of fish and d the number of days for the given growth period.

The effects on weight and length of temperature on the instantaneous rates of growth were evaluated using ANCOVA (covariance analysis), which was performed for linearized exponential models. A one-way ANOVA (variance analysis) was used to compare water quality and condition indexes between different age (i.e. sampling day) groups. Additionally, a Tukey-Kramer multiple comparison test was performed to statistically differentiate significant mean values for body weight and length.

5. Final product´s analytical composition and sensory evaluation

Analytical composition

In each sampling, in order to know the quality of the grown-out fish through analytical composition studies, a single muscle sample was taken from each individual. These samples were analyzed for protein, fat, humidity and ash content to study the organoleptic composition of fish.

Pools of individuals were made according to the aforementioned biological samplings and they were differentiated by thermal regimes to see if temperature’s manipulation could possibly have consequences in the final product’s composition. Individuals were collected and minced in a food processor (IKA® M 20 universal mill, IKA 1603601, Germany). Total protein was determined according to the Kjeldahl method (AOAC 1975). For lipid content analysis a rapid method of total lipid extraction and purification, presented by Bligh and Dyer 1959 was applied. Humidity and ash were measured by gravimetry according to the methods described by AOAC (1975) in a Heraeus oven and Heraeus furnace (Heraeus, Germany). Fig. M.5 shows a set of pictures of the procedures taken for the analytical composition study.

Fig. M.5. Cod and salmon analytical working pictures at the laboratory: (A) whipping the muscle samples for lipid extraction; (B) spinning the whipped sample; (C) sample ready to analyze; (D) weighing sample for protein extraction; (E) digestor during the protein extraction method; (F) and (G) Heraeus furnace; (H) humidity determination oven; (I) Kjeldahl distiller with an automatic valuator for nitrogen and protein evaluation; (J) lipid extraction method.

Sensory evaluation by seafood experts – cod experiment

A sensory evaluation was performed by a seafood experts panel (8 people) to study the acceptability of the final product only for the cod experiment, in comparison with other cod products that already were available on the market. The comparison was made between 3 different origin cod: wild cod, Norwegian farmed cod, and cod from this study.

A total of 18 cod fish individuals (mean weight 2,000 ± 30 g) were received and prepared in the laboratory. Six individuals were locally grown fish, another 6 were farmed in Norway (courtesy of SINTEF, Fiskeri og Havbruk), and the last 6 individuals were wild skrei cod from the local fish market. All samples of cod were obtained 48 hours prior to the sensory analysis to guarantee the optimal conditions for sensory analysis. The whole fish, without eviscerating, was used in the case of the local experimental cod, whereas eviscerated fish was used for both wild and Norway farmed cod. Samples of 5 g were taken from the loin part of the fillets and placed in aluminum boxes coded with three-digit random numbers for the cooked fillet evaluation. The samples (4 x 2.5 x 1.5 cm) were oven-cooked at 180ºC for 4 min. Samples were presented sequentially and nameless. Eight seafood tasting expert panelists participated in the sensory analysis, which followed the methodology explained in Warm et al. (2000). The methodology was based on two well-differentiated parts: (I) raw product parameters evaluation (i.e. gills, general aspect, eyes and texture), and (II) cooked product parameters assessment (i.e. aspect, smell, flavor and texture). All these parameters were evaluated using as reference a table with an adjusted scale from 3 to 10, according to the methodology of Torry Advisory Note No.91 (Tables M.4. and M.5.).

Table M.4. Some quality aspects of fish and fish products, and the senses used to assess them.

Sense	**Aspect of quality**
Sight	General appearance and condition, size, shape, physical blemishes, colour, gloss, identity
Smell	Freshness, off-odours and -flavours, taints, oiliness, rancidity, smokiness
Taste	Freshness, off-tastes and flavours, taints, oiliness, rancidity, smokiness, astringency, the primary tastes of acidity, bitterness, saltiness, sweetness
Touch (by fingers and mouth)	General texture, hardness, softness, elasticity, brittleness, roughness, smoothness, grittiness, gumminess, fluidity, wetness, dryness, crispness, presence of bones
Hearing	Brittleness, crispness

Table M.5. Sensory score sheet for cooked cod flesh taken from gutted fish that have been stored in melting ice.

Odour	Flavour	Texture, mouth feel and appearance	Score
initially weak odour of sweet, boiled milk, starchy, followed by strengthening of these odours	watery, metallic, starchy; initially no sweetness but meaty flavours with slight sweetness may develop	dry, crumbly with short tough fibres	10
shellfish, seaweed, boiled meat, raw green plant	sweet, meaty, creamy, green plant, characteristic		9
loss of odour, neutral odour	Sweet and characteristic flavours but reduced in intensity	succulent, fibrous; initially firm going softer with storage; appearance originally white and opaque going yellowish and waxy on storage.	8
wood shavings, woodsap, vanillin	neutral		7
condensed milk, caramel, toffee-like	insipid		6
milk jug odours, boiled potato, boiled clothes-like	slight sourness, trace of 'off' flavours		5
lactic acid, sour milk, 'byre-like'	slight bitterness, sour, 'off' flavours		4
lower fatty acids (eg acetic or butyric acids), composted grass, soapy, turnipy, tallowy	strong bitter, rubber, slight sulphide		3

Data collected were analyzed with the the SensoMineR v1.08 package within the R 2.12.2 statistical software. Data were tested for median, average, standard deviation, minimum and maximum, and Wilcoxon test to measure the “origin” factor's effect.

Consumers test – cod and salmon experiments

AZTI is experienced in performing consumers test for a several food products including bread, vegetables, cookies and fish or seafood products. AZTI has a database of regular sensory testers. From that database were identified and recruited by phone or email 80 and 30 consumers for cod and salmon studies, respectively. The difference in the consumer number was due to economic reasons. These consumers had attended consumer tests before, being aware of the methodology and procedures undertaken. Consumers were selected based on two criteria: to be older than 18 years, and to be a regular weekly consumer of seafood products.

Since the fish of both experiments were fed by commercial manufactured pellets and fish quality is generally accepted to be modulated by nutrition (Kiron 2012), fish product quality was only compared between randomly selected samples from aquaculture (i.e. without specific characteristics such as farming country) and wild origin (in case of cod) (Fig. M.6).

In the cod experiment compared individuals were locally grown, Norwegian farmed cod, and wild individuals. In the salmon experiment, compared fresh salmon products were from two different production origins: locally grown (i.e. in the present study and not commercial) and Danish farmed salmon (i.e. 5 individuals were bought in the local market with their origin in a RAS company located in *Hirtshals*, *Denmark*).

All fresh samples were obtained and prepared in the laboratory 24 hours prior to the sensory evaluation; the fish samples were beheaded, eviscerated and filleted and stored at 4ºC until their cook. Sensorial analyses were undertaken according to the norm UNE. 8587:2009.

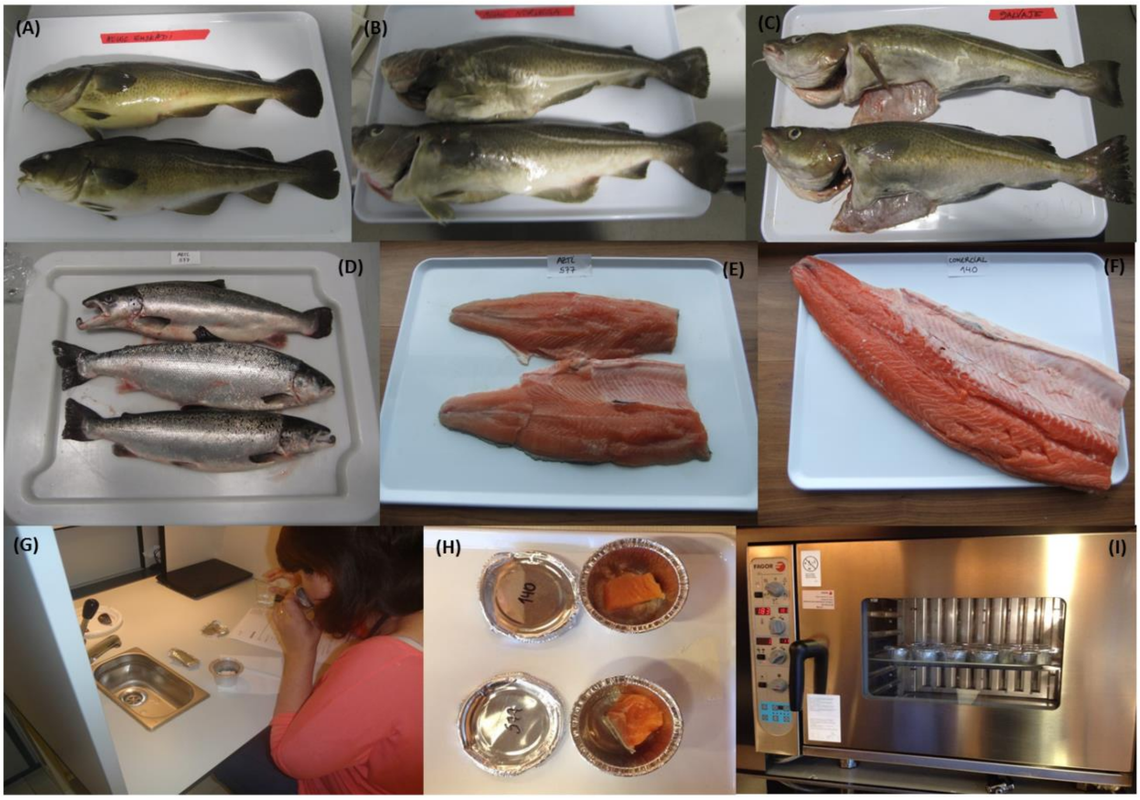


Fig. M.6. Cod and salmon individuals compared in the consumers tests: (A) locally grown cod; (B) Norwegian farmed cod; (C) wild cod individuals; (D) and (E) locally grown salmon individuals; (F) Danish farmed salmon; (G) a consumer tasting the product; (H) salmon samples prepared to be cooked; (I) samples in the oven.

Consumers were provided a questionnaire that asked them about their daily habits. The questionnaire was divided in two parts. The first section included questions on the consumer´s background and profile (i.e. gender, age and fish consumption), and the second on the acceptability of the tasted product during a blind-taste. The answers for each question were presented in 9-point hedonic scale to facilitate the work of the consumers in answering the test to describe characteristics of aspect, smell, flavour, texture and global impression: dislike extremely (1), neither like nor dislike (5), and extremely like (9). Consumers’ fish purchasing intention towards the farmed fish in the experiments was also evaluated through a structured 5-point scale ranged: definitely not buying (1), probably not buying (2), maybe yes/maybe no (3), probably buying (4), and, definitely buying (5).

At the beginning of both of the sensory tests, cod and salmon, samples were taken from the loin part of the fillets, and placed in aluminum boxes coded with three-digit random numbers for the cooked fillet evaluation. The samples, parallelepiped-shaped (3 x 2 x 2.5 cm) were oven-cooked (without steam) at 180ºC for 10 min. The samples were served directly from the oven to the consumers, and were presented sequentially and nameless. All tastes were individually carried out at individual and white booths. Each booth was previously prepared and supplied with: a napkin, a survey form, a pencil, a glass of water, some plain crackers, and an empty cup for discarding tasting samples. All the consumers received oral and written instructions, and they completed the testing at the same time and with no time limit. The sensorial analysis was carried out following UNE 4121:2006 methodology by quantitative response categorical scales. Data were tested for median, average, standard deviation, minimum and maximum, and Wilcoxon test to measure the "origin" factor´s effect.

Results

The results obtained within this thesis are presented in ***five Chapters***, each of them representing a different contribution.

Contribution 1: Land-based on-growing of Atlantic cod (*Gadus morhua*) using Recirculating Aquaculture System (RAS); a case study from the Basque region (Northern Spain).

Contribution 2: Land-based growth of Atlantic salmon (*Salmo salar*) and consumers' acceptance.

Contribution 3: Energy use in Recirculating Aquaculture Systems (RASs): a review.

Contribution 4: Integration of energy audits in Life Cycle Assessment (LCA) methodology to improve the environmental performance assessment of Recirculating Aquaculture Systems (RAS).

Contribution 5: Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges

Contribution 1

Land-based on-growing of Atlantic cod (*Gadus morhua*) using Recirculating Aquaculture System; a case study from the Basque region (Northern Spain)

This Contribution has been published in:

Badiola, M., Albaum, B., Curtin, R., Gartzia, I., Mendiola, D. (2016). Land based on-growing of Atlantic cod (*Gadus morhua*) using Recirculating Aquaculture System; a case study from the Basque region (Northern Spain). *Aquaculture* 468, 428-441.

Summary

Atlantic cod (*Gadus morhua*) is one of the most famous cold-water marine fish species. While the supply of Atlantic cod to European markets is relatively stable, this species continues to represent an interesting, and potentially lucrative, opportunity for commercial-scale aquaculture. Due to fishing restrictions and quota decreases over the last decade, the supply of Spanish-caught cod in European markets has declined, while the local demand for this species has increased. As wild-capture will not grow to meet this increased demand, land-based aquaculture of cod could represent a viable production model to satisfy demand. However, both capital investments and operation costs for a Recirculating Aquaculture System (RAS) farm are expected to be high, and therefore, all economic aspects must be taken into account before embarking on design or planning considerations. The influence of thermal control on growth, mortality and product quality also represent an important landmark when considering land-based aquaculture operations. The following represents a feasibility study to analyze the different economic scenarios and biological factors that can influence the business potential of growing this species in RAS. Two thousand five hundred cod individuals were reared at two different thermal regimes (i.e. controlled and natural range, respectively) through 2 pilot RAS set up in the Basque region (Northern Spain). The experiment lasted 430 days. Statistical differences were found in survival between different thermal regimes but no significant differences were detected within the fall or winter seasons. Daily specific growth rates were significantly different during the summer season with some compensatory growth patterns being observed in the natural thermal regime set up. Likewise, statistical significances were found between the fat contents from both temperatures after the summer period. Conversely, no significant differences were observed at sensorial level between the samples obtained within our pilot experiment and commercial samples from wild origin. Electricity use was found to be one of the most significant economic costs to be considered. The present study represents the first technical feasibility attempt on cod in land-based aquaculture from the north of Spain and demonstrates the technical feasibility to produce on-land based cod in the region, the equivalence of growth patterns with previous studies, the usefulness of the proposed thermal regime management as a tool for this species production, and the key economic parameters and thresholds for a potential feasible commercial activity in the region.

1. Introduction

Atlantic cod (*Gadus morhua*) is one of the most famous cold-water marine fish species, both from an economic and a socio-economic point of view (Kurlansky 1997). In the wild, there are several distinct stocks of Atlantic cod distributed along both sides of the Atlantic Ocean, where individual fish can attain ages of 20 years, sizes of 160 cm and weights of 40 kg.

Annual commercial landings of Atlantic cod have declined over the last several decades, with some stocks experiencing commercial extinction (FAO 2014). While overfishing has played a major role in this decline (Esmark and Jensen 2004), other contributing factors include variations in ocean temperature, natural predation, and fluctuations in populations of prey fish including capelin (*Mallotus villosus*) (Murua et al. 2007). At present, the supply of Atlantic cod to European markets is relatively stable and provided by a small number of northern fishing nations (i.e. Russia, Iceland, Denmark, and Norway), which accounts for over two-thirds of the total catch quota of the region. Due to population declines in the cod fisheries near North America, there is currently no supply to Europe from this region (Murua, pers. comm.). Thus, as supply has plateaued while demand has increased, this species represents an interesting, and potentially lucrative, opportunity for commercial-scale aquaculture.

The first production of juvenile Atlantic cod occurred in Norway in 1886 (Svasand et al. 2004). Around the year 2000, the life cycle of the fish had been closed and full-cycle production of the species in hatcheries became possible, although this production remained based on wild-caught broodstock as opposed to captive-bred broodstock (Jobling 1988; dos Santos et al. 1993; Bjornsson 1999). In 2002, after identifying a viable market opportunity for cod in Norway, the UK, Iceland, Canada and the USA, the first commercial breeding program began in Norway (Rosenlund and Skretting 2006). In 2005, more than 300 aquaculture licenses were issued in Norway to produce cod, albeit under limits of 65 t of production per company (Standal and Utne 2007, Fitzgerald et al. 2013). At the same time, other countries including Canada, Scotland, Iceland, and the Faroe Islands began developing cod aquaculture industries. However, the total global production volume never exceeded 12,600 t (Fitzgerald et al. 2013).

Atlantic cod´s (*Gadus morhua*) relationship with the Basque region (in the north of Spain) began in 1670: intensive fishing of whales by the fishing fleets of several neighboring countries (e.g. Germany, the UK, and the Netherlands) forced the Basque fleet farther and farther afield. The Basque fishers eventually arrived in the North Sea, Iceland, Svalvard (Norway) and Canada (Labrador and Terranova), where the boats discovered abundant populations of Atlantic cod (Garay 1985). While historically Basque-caught cod was available in significant volumes, presently cod´s fishing contribution to the total income of the Basque fleet has significantly decreased due to legislative restrictions and quota decreases in the aforementioned grounds. Conversely, local cod fish consumption has increased over time, with current demand in the Basque region alone now estimated at 3,500 t with a market value of nearly 30 million euro (MAGRAMA 2014).

The cod aquaculture industry is limited by the supply of juveniles and their high production costs (Bjornsson and Olafsdottir 2006); as such, recently there has been great progress to develop a steady and secure cod juvenile production model, with many scientific investigations focusing on this objective (Svasand et al. 2004; Bjornsson and Olafsdottir 2006; Foss et al. 2006, Imsland et al. 2007a; Remen et al. 2008; Fülberth et al. 2009; Moran and Stottrup 2011; van der Meeren et al. 2011).

The principal grow-out method for cod aquaculture is marine net pens (Bjornsson and Olafsdottir 2006). While cod have been extensively studied for many years, most of the existing investigations have focused on particular experimental growing conditions, such as stocking densities (Lambert and Dutil 2001; Bjornsson and Olafsdottir 2006; Foss et al. 2006; Bjornsson et al. 2012), as well as thermal (Bjornsson et al. 2001; Bjornsson et al. 2007) or photoperiod treatments (Imsland et al. 2005a; Imsland et al. 2007b; Fülberth et al. 2009). Studies on the viability of cod aquaculture in RAS are limited: Lambert and Dutil (2001) reached weights above 1 - 1.5 kg during studies using semi-recirculation systems. Rosenlund et al. (2004) also reared cod from 192 g up to 800 grams in recirculation systems. To date, only Fülberth et al. (2009) has reported an attempt at rearing cod to market size utilizing Recirculating Aquaculture Systems (RAS).

Recirculating Aquaculture Systems (RAS) involve a variety of water treatment components designed to remove metabolic wastes and recirculate clean water into the growing environment. The effective operation of RAS, which are mechanically sophisticated and biologically complex, requires education, expertise and dedication (Dunning et al. 1998). Prospective operators of RAS need to know about the required water treatment processes, the components involved in each process, and the technology behind each component. Many commercial RAS operations have failed because of component failure due to poor system design and inferior management (Masser et al. 1999; Badiola et al. 2012). A thorough knowledge of system design, specification of technical components and general operations is a minimum prerequisite for the success of a RAS farm (Badiola et al. 2012).

Capital investment for the setup of a RAS farm is normally much higher than that of many other traditional aquaculture production systems due to the requirement for additional equipment and energy to treat water for reuse. While the water treatment processes allow the opportunity to maximize environmental conditions for fish growth, they also increase operational costs, and, the failure of one single component in the system has the potential to result in huge economics losses (Summerfelt et al. 2001). Therefore, the economic aspects must be taken into account before embarking on any other design or planning considerations (Bijo 2007). A feasibility study is generally conducted during the early stages of a project to analyze the different scenarios and factors that can influence the business potential and subsequent sustainability (economic, environmental and societal) of the endeavor (Amanor-Boadu 2009).

The present study on land-based cod production aims to analyze: (I) the influence and management of local thermal conditions on fish growth and mortality; (II) the quality and consumers' acceptance of the obtained fish product and; (III) the derived cost-benefit scenarios.

2. Material and Methods

The experimental design followed in this study (i.e. location, RAS description, procedures for the water sampling, biological sampling and final product´s analytical composition and sensory evaluation and statistical methods) has been already described in the “Experimental design” section. Target species and study-specific materials and working procedures are presented below.

Targeted fish (Gadus morhua)

Two thousand five hundred individuals of Atlantic cod fish were obtained from the company Fosen Aquacenter (Trondheim, Norway) and received on the 7th of April 2012 via lorry. The fishes were uniform and randomly distributed. The initial mean weight and length (±SD) were 70.9 ±20.34 g and 187.15 ±14.48 mm, respectively.

Experiment´s methodology

The experiment´s chronology was set in line with the natural seawater temperature regime of the region and divided into 3 distinct time periods (Fig 1.1). The first period extended from April 2012 to June 2012; all the individuals were distributed uniformly and randomly in one pilot-scale RAS unit (i.e. in 3 different tanks). The maximum water temperature during this period was 16ºC. The second period began in June 2012 and extended through the summer months. During this period fish were divided into two systems, one designated as a “control module” with a working temperature range of 9-16ºC (tanks labelled as C1, C2 and C3) and the other one designated as “natural module”, with a working temperature range of 9-21ºC (tanks labelled as N1, N2 and N3). With the conclusion of summer, the third period began and both of systems were again set to the same parameters as the first period.

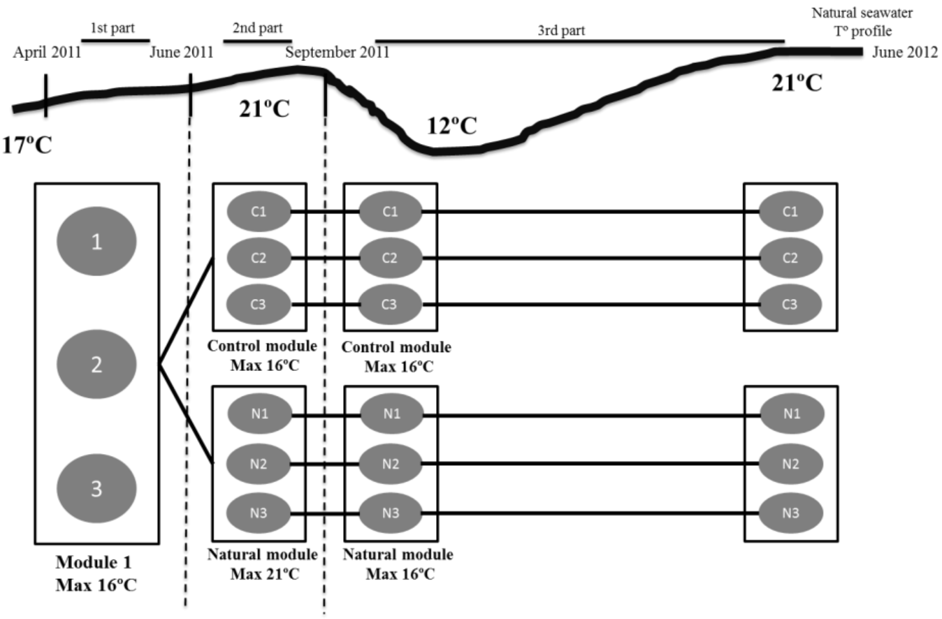


Fig.1.1. Experiment’s set up and chronology.

After 14 months of rearing, a brief trial was made to identify the thermal thresholds for mortality on the species; erratic swimming, lack of appetite or deposition over the tank bottom were observation keys to determine when the fishes were likely to be reaching the point of no return (PNR). Fishes were removed immediately to confirm a mortality event and to not impact the survival of their counterparts; all removals were registered, photographed and preserved. The thermal threshold experiment was concluded when 50% of the individuals per tank reached the PNR.

Initial stocking densities (9.73 kg/m^3), photoperiods (16:8 hours light: darkness) and dissolved oxygen (DO) concentrations (> 9mgL^{-1}) were similar at the beginning of the study in both modules. As the fish grew, those parameters were managed to keep them within the module's design limits, as well as to keep them comparable between modules. The fish were fed 3 times a day (8:00, 12:00 and 16:00) with a commercial floating dry feed (Europa 15F, Skretting S.A., Burgos, Spain) containing 56% protein, 16% fat and 13.4% carbohydrates. Pellet size varied according to the change in fish size; feed diameters ranged from 3 mm (at the beginning of the experiment) to 10 mm (at the end of the experiment). Daily feed rations were calculated depending on the cod biomass. Individuals were not treated with any antibiotics. Both the fish and the culture systems were visually checked on a daily basis to mitigate operation problems or mortality events. In case of any emergencies, an air stone was located at each tank to maintain the dissolved oxygen concentration near saturation.

2.1. Economic evaluation

The economic and biological data assessment that follows includes a comparison of: the described pilot scale scenario (including the control scenario) and 4 different simulated scenarios (noted later in this text as "SC."). The simulated scenarios represent products that would achieve three different levels of market price (high, medium, low): (I) pilot-scale production of 1,700 kg based on final average weights of 1.2 kg cod, (II) 60 Tn of production based on final average weight of 1.5 kg cod (SC.1), (III) 60 Tn of production based on final average weight of 3 kg (SC. 2), (IV) 200 Tn of production based on average weights of 1.5 kg cod (SC. 3), and (V) 200 Tn of production based on average weights of 3 kg cod (SC. 4). The criteria for the selection of these scenarios (as opposed to any other scenarios) were based on the local private interest for investment. Also, it is important to highlight that this analysis has not considered the timeline of Return On Investment (ROI). 3 cost categories were differentiated: initial investment, fixed costs and operational variable costs. First investment parameters are shown in Table 1.1.

Table 1.1 First investment (price in €) for the pilot-scale system and simulated scenarios (SC.).

	Pilot-scale	SC. 1	SC. 2	SC. 3	SC. 4	Method
Industrial land	19,451	275,000	275,000	770,000	770,000	Estimated[1]
Electrical infrastructure[2]	11,350	28,375	28,375	62,425	62,425	Per unit
Tank building[3]	13,600	293,564	293,564	645,840	645,840	Estimated
Biofilter maduration	288					Budget
Filters[4]		11,858	11,858	17,182	17,182	Budget
Total	44,689	608,797	608,797	1,495,447	1,495,447	

[1]: Industrial land for the experimental unit was achieved for 19,451 €. An estimation of 110 €/m^2 (data provided by the Urban Proyects database of the Housing and Social Issues Departments of the Basque Government) was used for simulated scenarios, including foundations, walls, land, roofs and infrastructures. 2,500 m^2 and 7,000 m^2 of land were estimated for the SC.1-SC.2 and SC.3-SC.4, respectively.

[2]: The electrical infrastructure was simulated according to the number of tanks. For SC.1 and SC.2 the number of tanks was assumed to be 10 and for SC.3 and SC.4 22.

[3]: Optimal stocking density requirements for cod (between 30-40 kg/m^3) were used in order to calculate the number of tanks for the simulated scenarios. SC.1 and SC.2, with a production of 60 T require 2,000 m^3 volume, and SC.3 and SC.4, 5,000 m^3. Average tank volume used was 236 m^3. 10 tanks were estimated for SC.1 and SC.2, and 22 tanks for SC.3 and SC.4. In the experimental scenario, tanks area was 20m^2 with a cost of 170 €/m^2. A simulated area of 173 m^2/tank, results in 29,356 €/tank.

[4]: Filters´ costs were given by a private company located in Spain. Drumfilters´ capacities were 6,000 and 14,000 l/h for the SC.1-SC.2 and SC.3-SC.4, respectively. At the same time, their costs were 9,800 and 14,200 €.

2.1.1. Assumptions for the different scenarios

Fixed costs included investment´s depreciation, equipment maintenance, electricity and insurance (Table 1.2). Depreciation (i.e. the investment´s loss of value over time) was estimated as the initial value of each element divided by its useful life.

Table 1.2. Fixed costs (price in €) for different scenarios.

	Pilot-scale	SC.1	SC.2	SC. 3	SC.4
Depreciation[1]	4,564	55,313	98,876	139,737	242,211
Maintenance[2]	3,691	57,739	100,240	135,174	237,834
Electricity[3]	1,003	1,003	1,739	1,003	1,739
Insurance	1,198	42,299	73,318	140,996	244,393
Total	10,441	156,054	270,573	416,776	724,177

[1]: Depreciation was calculated for each part of the investment: 20 years of operational life for tanks, 10 years for infrastructures and 5 for equipment.

[2]: Maintenance was separated between the building (1% of the value), infrastructures (5% of the value) and equipment (10% of the value).

[3]: Electricity contract was maintained between different simulations, so differences are due to the simulation duration.

Variable costs included energy, feed, oxygen, juveniles, water quality maintenance and salaries of the facilities' employees (Table 1.3). The system consumed an average of 29.40 kWh/kg (further explained in Badiola et al. 2017). Electricity cost was estimated according to input from professionals at the contracted power company. For SC.1, regular energy consumption rates were assumed, while during summer (i.e. second phase of the experiment when the cooling pump was in use) these rates were increased. In SC. 2, half of the mentioned energy consumption would be used for a longer period of time (26 months, instead of 14). SC. 3 and SC.4 increased this cost according to the longer production timeline associated with increased biomass production.

Table 1.3. Variable costs (in €) for different scenarios.

	Pilot-scale	SC.1	SC. 2	SC. 3	SC. 4
Used energy	7,659	104,305	90,398	347,684	301,326
Feed[1]	4,636	105,613	106,251	273,877	274,515
Oxygen	1,094	17,590	16,195	56,245	50,143
Salaries[2]	23,243	243,787	365,211	276,875	422,564
WQ maintenance	4,486	11,216	20,029	24,675	42,300
Juveniles	6,146	60,000	30,000	150,000	75,000
Total	47,264	542,512	628,083	1,129,356	1,165,848

[1]different FCR were assumed for different scenarios: 1.2 for SC.1 and SC.2, and 1.1 for SC.3 and SC.4.

[2] estimated with the average wage rate per year (company CEO, engineer and aquaculture technicians), in the Basque region. Number of employees varied depending on the production volume and product's marketable size.

With respect to feed, SC. 1 and SC. 2 required 72 t (assuming a FCR of 1.2 and a production of 60 t), while SC.3 and SC.4 required 220 t (assuming a FCR of 1.1 and a production of 200 t). Cost per kg of feed was 1.35€ for the first two scenarios and 1.15€ for the last two. Oxygen costs cover three parts: (I) used oxygen quantity, (II) cryogenic fee, and (III) the unloading cost. The cryogenic fee was fixed by the private fish company where the pilot scale modules were based (775.07€/month); so the authors assigned 5% of the total for this variable (38.75€/month). For the unloading quota, 5% (6.15€) of the farm´s total cost (122.97€) was assumed, and it occurred 6 times a year.

Finally, in order to estimate the returns, cod market price was calculated. Wild cod price is influenced by factors such as quality, freshness and time from harvest to reaching the market. On the contrary, aquaculture products have the advantage of a higher degree of control, with the opportunity to assure consistent availability and quality. Thus, farmed cod usually achieves a better market price than wild cod, however the final market price is also dependent on the individual´s size (i.e. bigger size equates to a higher price per kg) (FAO 2004). Three different price points were used for the analysis: (I) an average cod price of 3-4.50 €/kg, (II) a low price of 2-3 €/kg and (III) a high price of 4-6 €/kg. It should be noted that for bigger market size individuals (i.e. 3 kg or more), prices varied between 3 and 6 €/kg. Since the present study is focused on the first step of the value chain (fish production and first sale) of this species, the considered prices are for wholesalers (Fitzgerald et al., 2013). A summary of key assumptions taken for the economic evaluation is presented in Table 1.4.

Table 1.4. Key assumptions taken for the economic evaluation.

Assumptions	SC.1	SC.2	SC.3	SC.4
Target production (t)	60	60	200	200
Target market size (kg)	1.5	3	1.5	3
Nº fish input	50,000	25,000	170,000	85,000
Production cycle length (months)	14	26	14	26
FCR[1]	1.2	1.2	1.1	1.1
Feed cost/kg	1.35	1.35	1.15	1.15
Nº employees	3	5	4	6

[1] data based on the literature (Bjornsson et al. 2012; Colt et al. 2008).

3. Results

3.1. Water quality and growth performance

The experiment lasted 430 days. Water quality results are shown in Fig. 1.2. All parameters (except gas saturation) were successfully kept within appropriate ranges for correct survival and growth. The survival trends are shown in Figure 1.3. In both control and natural modules, mortality was observed to be significantly higher (ANCOVA, $P < 0.005$) during August (8.52 and 11.18%) and lower (ANCOVA, $P < 0.005$) during February (0.34 and

0.37%), respectively. During the fall and winter seasons, no significant differences (ANCOVA, $P > 0.05$) on fish survival were detected, and at the end of the study the cumulative survival rate ranged from 70% - 80% between the control and natural modules. The exponential equation provided a good fit for growth in length-at-age, explaining between 91 and 94% of the variability in L_t.

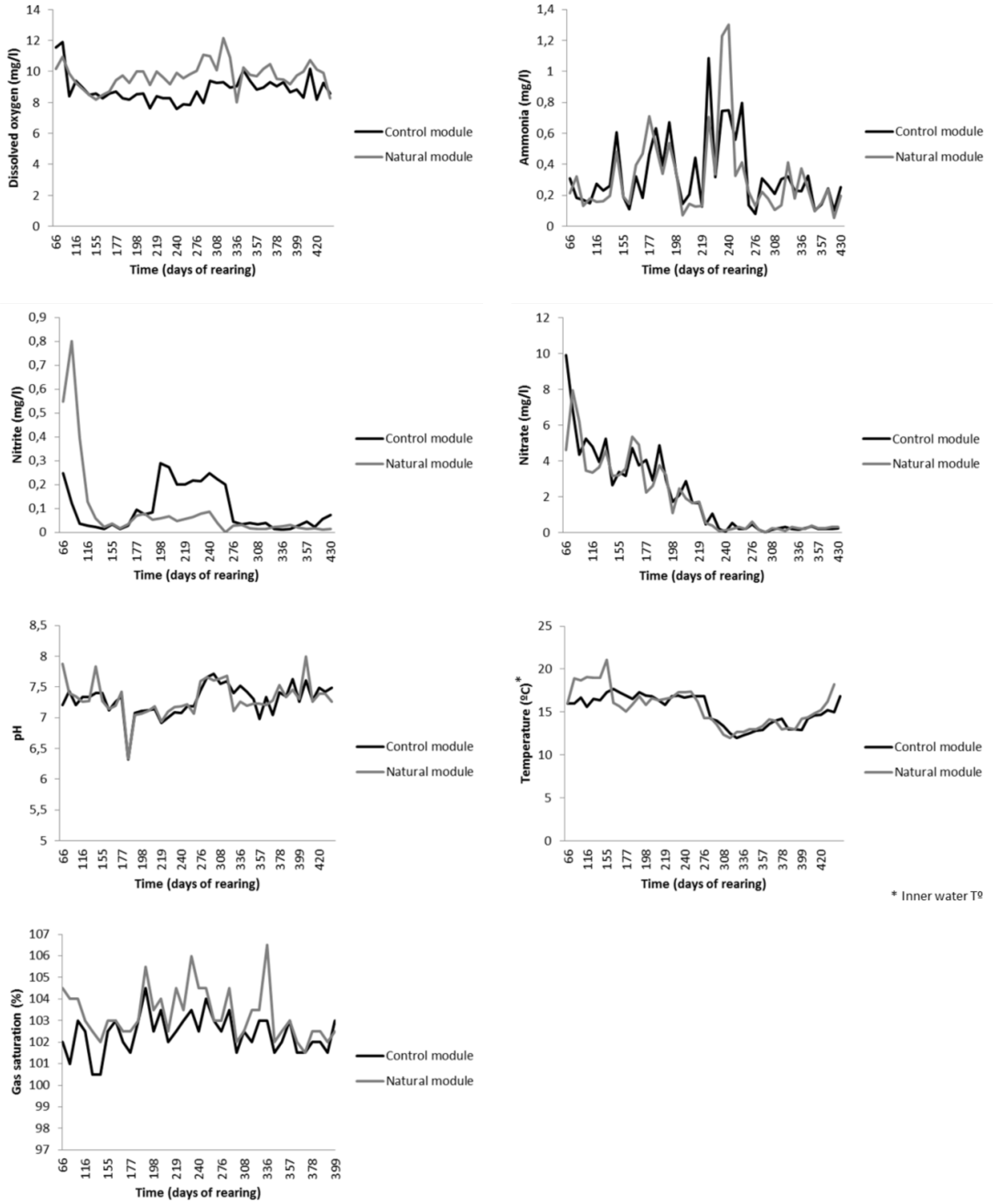


Figure 1.2. Physical and chemical parameters and their standard deviation, from beginning to the end of the experiment. Ammonia is expressed in mg l^{-1} NH_3-N (un-ionized), nitrite in mg l^{-1} NO_2-N and nitrate in mg l^{-1} NO_3-N.

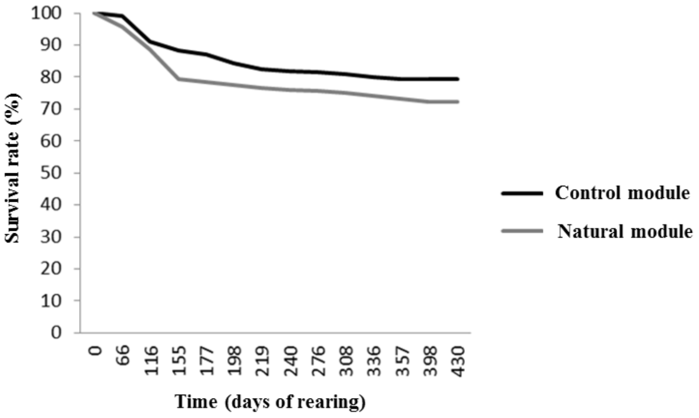


Figure 1.3. Survivorship curve (lines) taking temperature as a factor. The lines represent the observed surviving rate of cod (*Gadus morhua*) individuals.

For each module, the parameters of growth equations are listed in Table 1.5. Daily specific growth rates were affected by sampling time showing significant differences (ANOVA, $P < 0.05$) during the summer season (Fig. 1.4). Both condition and hepatosomatic indexes were also affected (ANOVA, $P < 0.05$) by sampling time. While time (i.e. season) had the most significant impact on daily specific growth rates, at the end of the study no significant differences (ANOVA, $P > 0.05$) were detected between individuals from different regimes. At the natural module, lower mean values were found in summer (June 2012) and the highest values from the control module were also found during the same dates. From winter 2013 onwards, both indexes showed compensatory recovery and were kept stable until the end of the study; no significant differences (ANOVA, $P > 0.05$) were found between the final index values from both culture modules. Food Conversion Ratio (FCR) was 2.34 ± 0.50 and 2.09 ± 0.65 at control and natural modules respectively. No significant differences in FCR were found ($P > 0.05$) at the end of the experiment.

Table 1.5. Growth values: (A) Estimated parameters of the length-at-age exponential growth equation for each thermal experiment. Equations are given in text. (B) Instantaneous (G), relative (K) and daily specific (%) growth rates, of weight, calculated from the equations reported by Ricker (1975). Statistical significance (*) was set up at (p < 0.05) (pairwise comparison).

Control module (9-16ºC)					**Natural module (9-21ºC)**				
W0	**G**	**K %**	**Nº observations**	**R^2**	**W0**	**G**	**K%**	**Nº observations**	**R^2**
70.9	0.006	0.599	239	0.962	70.9	0.0059	0.592	242	0.95
L0	**G**	**K%**	**Nº observations**	**R^2**	**L0**	**G**	**K%**	**Nº observations**	**R^2**
18.71	0.0018	0.184	239	0.91	18.71	0.0018	0.181	242	0.94

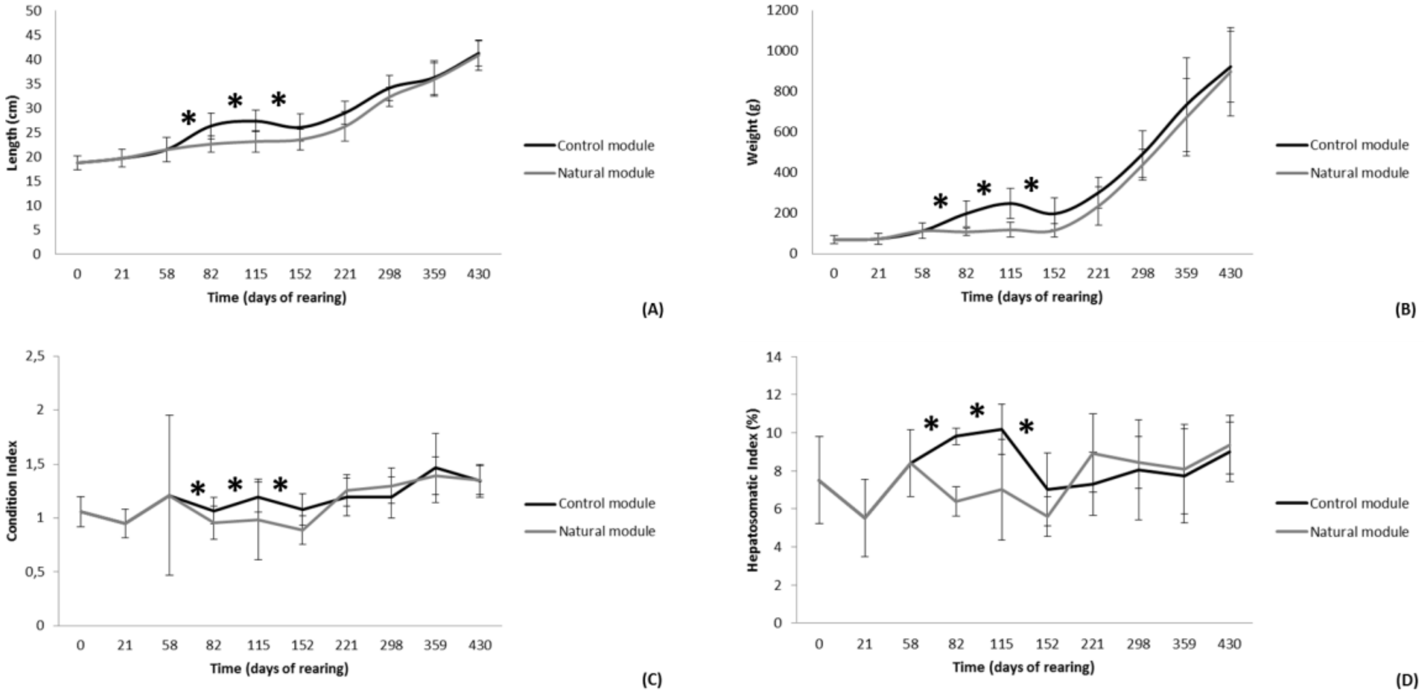


Figure 1.4 Atlantic cod (*Gadus morhua*). Change in mean (± SD): (A) body´s length, (B) fish weight, (C) condition index, and (D) hepatosomatic index. Lines represent mean (± SD) in control and natural module (black and grey lines, respectively). * represents significant pair wise comparison test (P < 0.05).

3.2. Final product´s analytical composition and sensory evaluation

3.2.1. Analytical composition

The results of the proximate analytical composition (presented as fat, humidity, protein and ash content) are shown in Fig. 1.5. Significant differences ($P<0.05$) were found in terms of fat content after the summer period between control and natural modules. The natural module suffered a loss in fat content after being at high temperatures although this was recovered and eventually surpassed at the end of the experiment. In terms of protein, humidity and ash, no significant differences ($P > 0.05$) were found between the two modules.

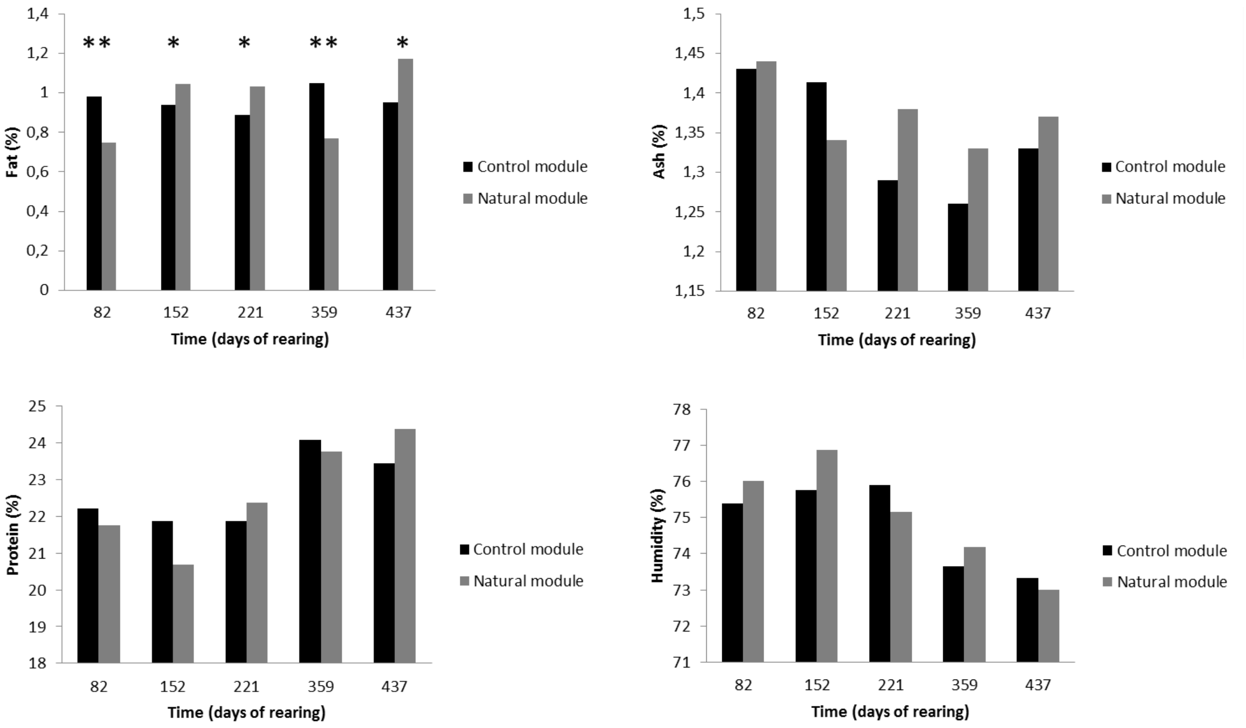


Figure 1.5 Results for the proximate analytical composition, presented as fat, humidity, protein, and ash content for both of the Tº regimes. Significant differences are shown with * or **.

Fat content was found to be significantly higher ($P < 0.05$) in the cod individuals reared from the present study (0.95% in controlled Tº cod and 1.17% in natural Tº cod) as compared to the wild cod (0.45%) or the farmed cod samples (0.37%) utilized in the final product comparison. The highest protein content was also higher ($P < 0.05$) in individuals from the experiment (23.43% in control module cod and 24.37% in natural module cod), followed by cod farmed in cages (20.87%) and wild cod (20%), respectively. The samples of wild cod and cod farmed in cages presented similar ($P > 0.05$) humidity contents, 77.62 and 77.78% respectively, while cod from the presented RAS experiment resulted in smaller percentages (73.32% for control module cod and 73% for natural module cod). Regarding ash content, RAS-reared cod presented the highest content (1.37% natural module cod and

1.33% control module cod), 1.18% in wild cod and 1.07% in cod farmed in cages; however, differences were not statistically significant.

3.2.2. Sensory evaluation

Cooked cod

Several qualitative differences between wild and farmed cod were observed during the sampling of cooked product in the present study. With respect to the wild cod, descriptions included odors and tastes of boiled seafood, in addition to an herbal and metallic taste; compared to farmed cod, which was described as neutral in flavor. With reference to the product texture, the individuals reared in the presented study showed the most solid characteristics as compared to the products from the other two different origins.

Raw cod

Descriptive words given by the experts for the raw cod are shown in Table 1.6.

Table 1.6. Descriptive words given by the experts during the sensory evaluation. Experts followed the Torry Advisory Note nº 91 - Sensory Assessment of Fish Quality (Annexe A), where there are suggested words for general appearance, gills, eyes and texture for raw fish, and smell (odour), taste (flavour) and texture for cooked fish.

RAW COOD	Appearance	Gills	Eyes	Texture	Score
Cod farmed within the present study (RAS)	Brilliant skin, skin colour dark green moss with green olive colour spots, well-marked and whitish lateral line, lightly yellowish ventral area, dark-red tail.	Well-defined red-purple colour prints, without mucus, neutral smell (just cutted grass), metallic, marine	Deep-set, concave, white pupil (some gas problems), dull cornea, small eyes	Very firm	9-8
Norway farmed cod (cage farming)	Moderated brilliant skin, grey-green-brownish colour, Grey-greenish, thick lateral line, white well-marked, greenish fins, grey-greenish tail. In general, white-yellow color	Brown-beige color, moderated and viscous mucus, laminated and fattened, not well defined, hay odour, grass, yeast, rancid, slightly acid	Flat, whitish cornea, slightly turbid, dark pupil	Firm, light rigidity loss	9-8
Wild cod	Moderated brilliant skin, grey green-brownish color, grey-white color belly and grey ventral fins. Dorsal line marked. Generally, deep-red grey color with green-brown spots	Brownish colour, dark brown, abundant and viscous mucus, fatty prints and not well defined, hay odor, grass, old rancid	Flat or slightly deep-set, translucent cornea and lightly muddy, black pupil, opalescent cornea	Firm texture, some loss of rigidity	9-8

COOKED COD	Smell	Taste	Texture	Score
Cod farmed within the present study (RAS)	Boiled milk intense, starch, lightly vegetal, neutral	Neutral, sweet at the end	Very solid, solid, solid. Soft, juicy, succulent, quite gummy at the end	9-8
Norway farmed cod (cage farming)	Dairy, boiled milk, slightly sweet	Neutral, sugary, slightly acid, bitter at the end	Firm, dry, slightly dry, gummy, it is easily crumbled in different layers (crumbly)	9-8
Wild cod	Softly to boiled seafood, cooked fish, vegetable, salty but sweet at the same time	Characteristic from cooked cod, cooked seafood with an herbal touch, lightly bitter, neutral	Soft texture, not consistent, fibrous, juicy, succulent, tender, crumbly	9-8

3.2.3.Consumer´s test

With respect to the demographic characteristics of the consumers, 63% were female and 37% were male. Regarding age, 43% were 25-34 years old; 29% were 35-44 years old; 16% were 45-54 years old; 10% were 55-64; 1% was 18-24 years old and 1% > 65 years. With respect to their fish consumption habits, 69% consume fish once a week; 30% once per month and 1% more than once a week.

The results of the consumers' evaluation on aspect, smell and flavor are shown in Table 1.7 and Figure 1.6.

Table 1.7. Average, minimum, 1st quartile, median, 3rd quartile, maximum and Wilcoxon test values for origin factor.

	Average	Minimum	1st quartile	Median	3rd quartile	Maximum	Wilcoxon test	
							W	p-value
APPEARANCE								
Farmed cod	6.620	4	6	7	7	9	2282.0	0.308
Wild cod	6.775	1	6	7	7.5	9		
SMELL								
Farmed cod	6.127	4	5	6	7	9	2483.0	0.876
Wild cod	6.113	2	5	6	7	9		
TASTE								
Farmed cod	6.310	2	6	6	7	9	2612.0	0.703
Wild cod	6.169	2	5	6	7	8		
GLOBAL SCORE								
Farmed cod	6.282	3	6	6	7	9	2377.0	0.545
Wild cod	6.359	2	6	6.5	7	9		

The results did not show any significant differences (P > 0.05) at a sensorial level between the samples from aquaculture and wild origins. Thus, the global score for both of the products (i.e. farmed and wild cod) were similar (6.282 and 6.359, respectively), with no significant differences (P > 0.05).

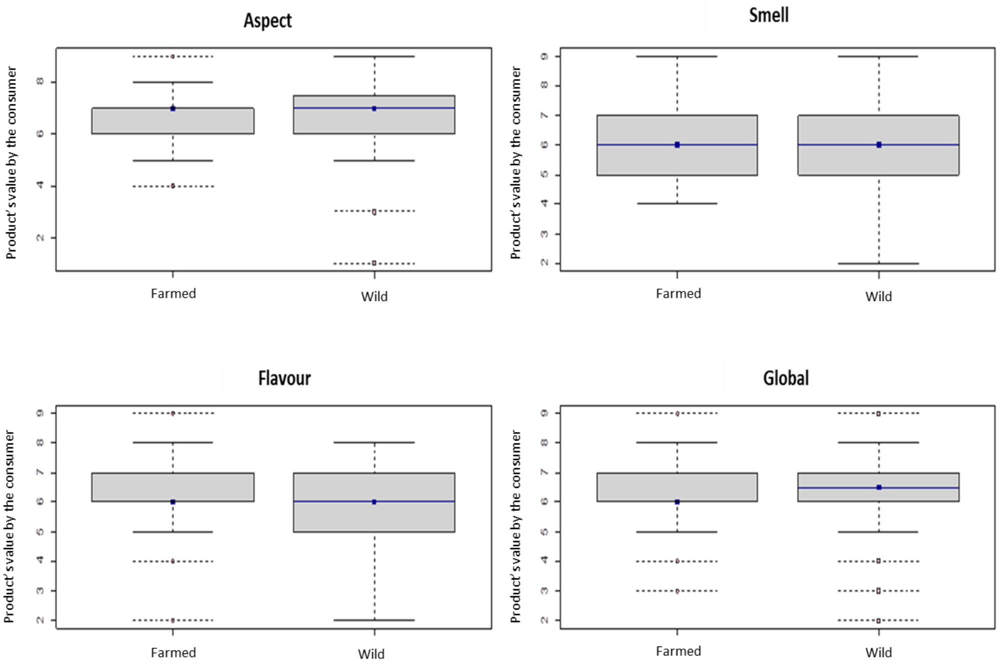


Figure 1.6. Consumers evaluated aspect, smell and flavour for two different origins: wild and farmed cod; average, minimum, 1st quartile, median, 3rd quartile and maximum are shown.

3.3 Economic evaluation

Employees' salaries (40% of the total cost of production), followed by electricity use (13% of the total cost of production) and juveniles (11% of the total cost of production) were found to be the most significant economic costs within the experimental framework. Costs per unit of product are shown in Fig. 1.7. As observed, fixed costs increased with the production volume, with maintenance and insurance representing the largest proportions. Within variable costs, which are directly dependent on production volumes, energy use and feed were shown to represent the largest costs. Calculated operational cost/kg vs. price/kg are shown in Fig. 8 from each of the experimental scenarios. As observed, benefits did not overtake costs in any of the estimated scenarios, however it is noteworthy to highlight how the benefits are significantly increased in SC.2 and SC.4, when the production is simulated for 26 months. The calculated equilibrium points (i.e. the production volume necessary to ensure profitability) for each proposed RAS scenario is shown in Table 1.8.

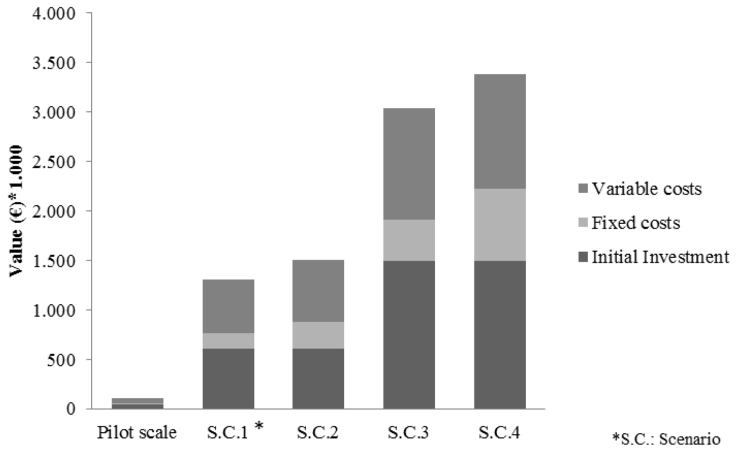


Figure 1.7. Costs per unit of product.

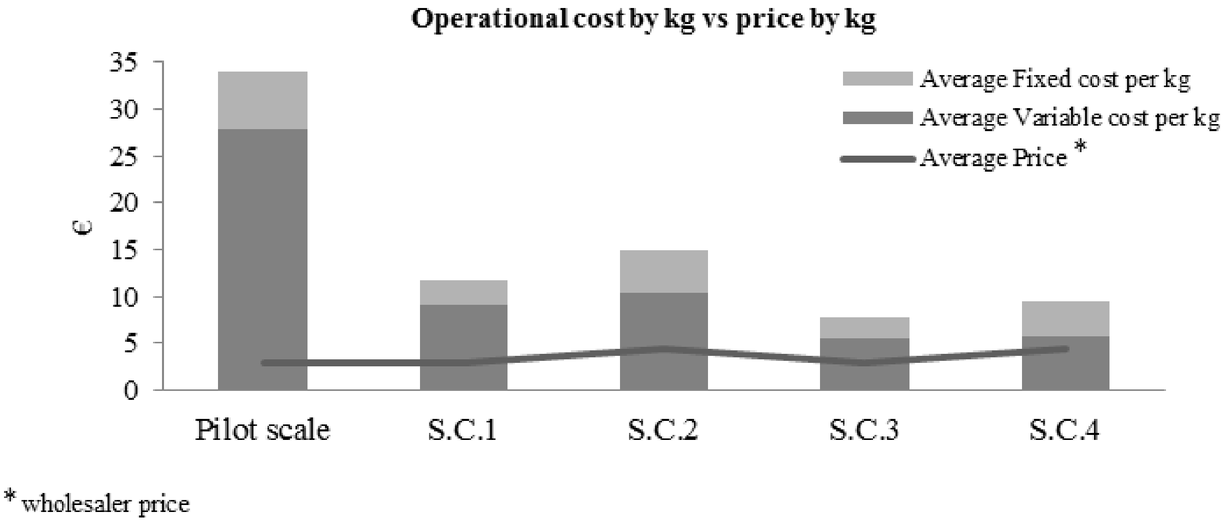


Figure 1.8. Operational costs depending on production volume (kg) and market price per kg of cod.

Table 1.8 Equilibrium point for each of the proposed RAS scenarios for cod rearing within the present study.

Estimated production (T)	Marketable size (kg)	Price per kg (€)	Production´s Equilibrium Point (T)
60	1.5	2	349
		3	233
		4	175
	3	3	300
		4.5	200
		6	150
200	1.5	2	774
		3	516
		4	387
	3	3	631
		4.5	420
		6	315

4. Discussion

Cod is one of the most popular and commercially valuable marine finfish species for human consumption in Europe. Over centuries of active trade, there has emerged a significant and diverse market at several different levels and niches, from commodity to premium (Fitzgerald et al. 2013). Thus, there exists a large, complex and well-developed market for a range of cod forms and products. One premium example is cod liver oil, a high-value human nutritional product used to supplement omega-3 fatty acids, (i.e. eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA)) considered essential to prevent certain health issues.

In the Basque region, RAS represents the only technology available for large-scale commercial development of cod aquaculture (EJ-GV 2008). It is generally accepted that such RAS technology offers potential advantages for aquaculture development including the ability to place the farms in locations where water resources are limited and/or near to the market, where product transport time and costs can be reduced (Hutchinson et al. 2004). In this context, carrying out a feasibility study, in which all biological, economic and product quality aspects are considered, represents an appropriate and necessary endeavor prior to initiating investment in commercial scenarios.

While there are cod-focused RAS studies that have been conducted in Northern Europe, these studies focus mainly on post-hatch larvae and early life stages (e.g. Pedersen and Jobling 1989; Otterlei et al. 1994; Knútsson 1997; Lambert and Dutil 2001; Björnsson and Steinarsson 2002; Foss et al. 2004; Mokness et al. 2004; Bjornsson and Olafsdottir 2006); to date, no study had been published in Southern Europe regarding land-based aquaculture of Atlantic cod (*Gadus morhua*) to marketable size. Thus, the results of the present study represent the first available data on the growth, condition, biochemistry, economics and product quality of Atlantic cod (*Gadus morhua*) cultured within a land-based scenario in the Bay of Biscay.

In the present experiment, both growth regimes (i.e. control and natural modules) showed acceptable water quality and mortality rates, albeit some differences in growth performance were found. Biofiltering systems in both regimes performed adequately, as evident by the ammonia concentration staying lower than the threshold limit established by Foss et al. (2004) and Bjornsson and Olafsdottir (2006). This was achieved by controlling feeding and fish stocking density, as well as maintaining oxygen concentration above saturation levels. Some authors (e.g.Thruston et al. 1981; Wajsbrot et al. 1991; Foss et al. 2007 and Remen et al. 2008) have already reported that cod is more tolerant to ammonia under mild hyperoxia conditions. Conversely, lower oxygen levels lead to appetite reduction and growth depression (Whitworth 1968; Jobling 1994; Chabot and Dutil 1999; Timmons and Ebeling 2010). While significant differences in growth performance were found during the non-favorable thermal period, suitable water quality parameters were maintained during the whole experiment. While in general nitrite concentrations were maintained below the threshold limits reported (e.g. Siikavuopio and Sæther 2006) one nitrite peak did occur from Day 66 to about Day 155, matching an increase in water temperature and a slight decline in survival rate during this time. Likewise, the consistent reduction in nitrate concentrations as the experiment progressed supports the adoption of a partial water reuse modulation strategy. This strategy was mostly developed to take advantage of the seasonal suitable thermal conditions (i.e. < 16ºC; from late autumn to late spring); these conditions should be understood as minimum requirement to carry out any feasible cod production in the Basque region. While the aforementioned procedure can be used at an experimental scale, further investigations should be developed before proposing any commercial initiative.

Gas saturation represented a concern during some periods in the present study. Saturation levels detected in the seawater (average between 102-103%) resulted with some fish suffering from bubbled eyes and apparent cataracts towards the conclusion of the experiment. Previous studies examining possible causes of similar issues (i.e. bubble eyes, erratic swimming) referred to chronic gas saturation exposure as one of the main causes (Gunnarsli et al. 2008; Moran and Stottrup 2011; Moran et al. 2012). The authors of the present study suspect that the origin of these health issues could be related to engineering designs. Thus, some designing aspects were carefully modified at the end of the experiment and helped to decrease daily levels of gas saturation: for example, (I) several barriers were set up in the expansion tank to decrease the impact of the entering water and; (II) distances between the growing tanks and the expansion tank were increased to lower water velocity in this section. In general, special attention should be taken into account regarding gas concentration levels when working with RAS.

During the present study, a compensatory growth (Jobling 1994) was observed within the natural module. Luczkovich and Stellwag (1993) and Imsland et al. (2005b), also showed that early environmental (i.e. Tº) manipulation towards colder water could lead to a long-term positive growth effect in Atlantic cod, and this effect may stimulate benefits in commercial aquaculture (e.g. faster growth in a given timeframe). Hanson (1996) and Björnsson and Steinarsson (2002) agreed that fish should be reared at stepwise temperature regimes, mimicking natural oceanic conditions and fluctuations. Likewise, Bjornsson et al. (2001) reported growth enhancement when Atlantic cod must adapt their enzymatic activity to different environmental conditions. During the present study, the natural module experienced higher temperature conditions during the summer period, resulting in a very quick recovery and higher growth rates at the end of the experiment (after 485 days). Bjornnsson et al. (2001) concluded that slower growth rates are experienced in cod fish as the size of the individual increases, and indicated that 17ºC and 7ºC were the proposed optimum temperatures for growth at 2 g and 2 kg respectively. Pedersen and Jobling (1989) also stated that larger fish grow better at lower Tº and set the ranges of 11-15ºC and 9-12ºC for individuals below 1,000 g and over 1.5 kg, respectively.

As shown in the present study, the individuals from the natural thermal regime (i.e. natural module) experienced a halt in growth during the summer period when warmer water temperatures were present. The warmer water was shown to decrease individuals’ appetite (i.e. feed consumption was decreased during high temperature season), arresting growth without leading to mortality. Their swimming behavior was observed to be slow (probably as a strategy to keep energetic expenditure to the minimum biological requirement levels) and they mostly remained at the lower part of the tanks. Thus, as reported by Jobling 1994, fish and other animals are able to adapt to feast-and-famine conditions by showing marked growth spurts when environmental conditions and food supplies are increased after a period of starvation. Moreover, it is normally the fish that are in the poorest physical condition that show the greatest response, displaying immediate mortality or the most rapid rates of weight gain when adequate rearing conditions are restored. Jobling (1994) also reported some cases of complete somatic growth recovery from starvation.

Liver weight and both the condition and hepatosomatic indexes also indicated a growth regression during the summer period. The repletion showed a rapid increase in the weight of the muscle, a relative increase in muscle lipid and protein content and a corresponding decrease in the % of muscle water. This is consistent with previous findings by Jobling (1994). Likewise, the rebound in food conversion ratio (FCR) and rate of growth (g day^{-1}) indicated that the physiological recovery of the individuals reared in the natural module (i.e. the stepwise Tº regime) had been completed.

Fish quality has been defined as “a combination of such characteristics as wholesomeness, integrity and freshness” (Martin 1988). Organoleptic properties and nutritional value are two sets of characteristics that, together with freshness, represent those qualities comprising fish quality as perceived by the consumer. Some authors (e.g. Huss 1988; Grigorakis 1999) have reported that organoleptic properties and nutritional value both strongly depend on the chemical composition of the fish, which in turn depends on a variety of different factors including intrinsic (i.e. species, genetics, age, sex etc.), environmental (i.e. temperature, salinity etc.) and feeding factors (i.e. diet composition and feeding ratio).

As shown in the present study, organoleptic composition and tastings of cod product resulted in positive feedback from the sampled consumers. Descriptive analysis is undoubtedly one of the most valuable sensory tools to provide detailed information on product quality and specific properties (Murray et al. 2001). Organoleptic tasting revealed that the quality of cod fish reared over 14 months in RAS showed equal or similar characteristics to the comparative samples obtained from the fish market. The panelists were not able to discriminate between samples or origins (i.e., aquaculture and wild) prior to sampling, and the resulting scores were almost equal across the board with respect to smell and taste attributes. Further, the panelists identified a firmer and more attractive texture in the farmed cod than in the wild samples. All these results indicate the consumers´ acceptability of the products tested.

The most critical factor of RAS farmed fish is an odorous smell produced by two chemicals: geosmin and 2-methyl-iso-borneol; these compounds are implicated in the earthy/muddy off-flavours present in many farmed fish (Howgate 2004). However, no unpleasant smells were ever reported by the panelists in the present study.

In general terms, the results showed that Atlantic cod can afford seawater temperatures of 21ºC for a maximum of 2.5 months and still complete the rearing process to commercial size, if all environmental conditions (feeding, density, temperature) are correctly restored after this time. The present study suggests the importance of having efficient thermal control systems available in the RAS market. As mentioned, thermal regime management can display immediate mortality or more rapid rates of weight gain with no significant statistical impact on FCR, but it may also influence final product's qualities (i.e. higher fat and protein contents, as resulted in the present study).

Regarding economics, as calculated and simulated herein, the marketing and selling of 200 t of cod at 4.5 €/kg would provide a total revenue of 900,000 €. This estimated cod price is equal to that of the Norwegian wild cod utilized in this study. However, there is a high degree of uncertainty regarding the realistic expectation of achieving this price in the study region (i.e. the Basque country). One study from Ireland concluded that the market price for farmed cod products had been high and relatively stable during the years preceding 2009 (Fitzgerald et al., 2013); the same authors note that one ton of farmed cod was worth 4,000 euros in Norway. The price had to decrease rapidly at the beginning of 2009 with the recovery of commercial Atlantic cod fisheries and the resulting fishing in NAFO areas (González-Costas et al. 2013). In this way, the current accepted domestic price of cod fish in the southern market is 2.91 €/kg. Furthermore, feed price (i.e. its ingredients price) and the quantity used while farming (i.e. FCR) are also important parameters dictating the economic viability. In the present study, due to a lack of data about commercial-scale RAS cod farms' FCRs, the following examples were taken as reference: (I) a study made by Bjornsson et al. 2012, where it was presented that FCRs range from 0.81 to 1.02 in juveniles cod (from an initial weight of 44 g until 242 g, the biggest individuals) and; (II) a study made by Colt et al. 2008 where the calculated farming of salmon with FCRs was reported as 1.1.

Appropriate management is also shown to be invariably important to successful operations; when management measures are appropriate, lower FCRs may be achieved as less amount of feed is wasted (i.e. knowing fish behavior). Therefore, although it is generally accepted that RAS offer advantages in locations where there are limited resources or close to market, all direct local variable costs (such as energy, feed, salary, etc.) will greatly affect both the final product price and local commercialization.

When compared with wild cod, farmed cod presents several benefits to the producers: for example, a price increase of 20% for fresh product forms as well as the opportunity for consistent supply to the market throughout the year (FAO 2012). However, the assessment of cost benefit at the global scale is uncertain and complex; it must reflect not only the production factors but also the market needs/opportunities, the local consumer demand, the effects of management competency of producers and the context of economies of scale.

5. Conclusion:

Aquaculture involves not only animal husbandry (biology), but also engineering (the technology used), markets' acceptance of the final product (society/culture), associated environmental impacts (environment) and financial costs (economy). Moreover, commercial aquaculture is criticized for the associated environmental impacts, which provided the motivation for this feasibility study and stresses the need to study each industrial activity in detail.

The present study has combined different research methodologies to assess feasibility of a particular aquaculture case-study. Albeit, both biological and product quality approaches were satisfactory, higher dimensions of simulated economic scenarios should be properly considered within future studies. Within the present study, an economy of scale framework is not illustrated and subsequently shows that pilot-scale research models are costly and risky, leading to the general acceptance that their main role is to improve foundational understandings and contribute to applied scientific knowledge.

The study concludes that land-based scenarios producing less than 200 t of cod fish may not be economically viable in a geographic zone where both salary and energy costs are limiting factors. Therefore, a clear dimension and perspective of economies of scale should be considered if affordable operational costs and consistent marketable final product prices are intended.

Contribution 2

Land-based growth of Atlantic salmon (*Salmo salar*) and consumers' acceptance

This Contribution has been published in:

Badiola, M., Gartzia, I., Basurko, O.C., Mendiola, D. Land-based growth and sensory evaluation of Atlantic salmon (*Salmo salar*) in Northern Spain: looking consumers' acceptance. *Aquaculture Research* (accepted, DOI expected for Sept. 2017)

Summary

Atlantic salmon (*Salmo salar*) is currently the highest-valued species grown in Europe. The industry has been on the frontline of public concerns regarding sustainability which has increased the use of Recirculating Aquaculture Systems (RAS). Salmon has changed from a luxury product to a global commodity. Nevertheless, food products need to meet consumers demand for the industry to be successful. Descriptive sensory tests present a sophisticated tool for the comparison of product prototypes to understand consumer responses in relation to sensory attributes. Aquaculture is being promoted in the Basque region with the aim of creating a sustainable and complementary economic activity to the fishing and seafood sectors; with a priority given to RAS and salmon as a potential technology and species, respectively. Compensatory growth using local seawater temperatures, and consumers' final product acceptance and purchasing intention through a hedonic evaluation were studied. One thousand five hundred salmon individuals were grown for 497 days at two different thermal regimes in two pilot-scale RAS units. Growth rates were significantly different for both temperature regimes during the second summer season with some compensatory growth patterns being observed along the timing of the natural thermal regime set up. Conversely, no significant differences were observed at sensorial level between the fillet samples obtained in this study and commercially grown RAS salmon from Denmark. Consumer level of acceptance and product purchasing intention reflect the possibility of marketing RAS grown salmon in the local markets. The present study refers the first technical attempt on salmon land-based aquaculture from Northern Spain.

1. Introduction

The salmon farming industry has grown substantially in the past 40 years, coinciding with the decline of wild stocks (Gross 1998). In 1996, salmon aquaculture overcame the fishing products industry as the most important supplier of salmon products worldwide. By 2004, global production of farmed salmon exceeded its wild catches by more than one million metric tons (hereafter, t) (Asche et al. 2013; NASCO 2014; FAO 2016). A decade later, in 2014, the excess was doubled (i.e. 2,326,288 t salmon were produced, while 2,319 t were caught from fisheries) (FAO 2016). Currently, salmon aquaculture is considered the fastest growing food production system in the world, accounting for 70 percent (i.e. 2.4 million t) of the market (Shepherd and Little 2014).

In Europe, Atlantic salmon (*Salmo salar*) is the highest-value species accounting for 21.32 % of the intra-communitarian economic value of fishing products (COM 2016). Production of Atlantic salmon smolts (i.e. individuals of certain size and age reared in hatcheries to be grown thereafter in sea cages or land-based facilities to marketable size) in Northern Europe takes place mostly in Norway, Scotland, the Faeroe Islands, Iceland and Ireland. Norway, the leading producer, accounts for some three quarters of the total annual production (Bergheim et al. 2009), with an annual output close to 250 million smolts. Scotland represents the largest salmon producer country within the EU, with an output of 14 t in 1971 and 163,000 t in 2013 (Munro and Wallace 2015). Ireland became an important Atlantic salmon producer for the European and US market in 1980. However, the industry was not developed and the production dropped from 25,000 t in the 1990s to around 10,000 t in 2015 (Warrer-Hansen 2015).

Salmon farming industry has been on the frontline of public concerns regarding sustainability and it has attracted criticisms and a preponderance of bad press (Naylor and Burke 2005; Amberg and Hall 2008; Shepherd and Little 2014). Examples of criticisms are the magnitude of discharge of nutrients, organic particulates and chemicals (Buschmann et al. 2006) along with , types of pathogens, and escapee interactions with wild stocks (Dempster et al. 2002; Buschmann et al. 2006, Uglem et al. 2014), culling of predators, and use of industrial fish in feed (Naylor et al. 2000). Additionally, the environmental restrictions for fish farming have increased in many countries (Fernandes et al. 2001). In this context, the use of Recirculating Aquaculture Systems (RAS) has increased as a new technological solution to provide sustainably farmed fished. Such systems offer several advantages over traditional net-pen systems, such as reduced water consumption (Verdegem et al. 2006), improved opportunities for waste management and nutrient recycling (Piedrahita 2003), better hygiene and disease management (Summerfelt et al. 2009a; Tal et al. 2009), and control of biological pollution by fostering no escapees (Zohar et al. 2005).

The presence of RAS is growing in the salmon industry. Available data suggest that hatchery production system (i.e. nursery production of smolts) has already moved from its original operation practice (i.e. flow-through or small sea cages) towards the intensive use of RAS technology due mainly to: (I) avoid performance issues (e.g. lordosis) during pre-growing (Bergheim et al. 2009); (II) minimize the risk of water quality and supply (Joensen 2008; Kristensen et al. 2009); (III) avoid seasonal seawater thermal variability (Kristensen et al. 2009); and, (IV) secure growth, survival and smolts quality (Terjesen et al. 2008; Martins et al. 2010). In the Faroe Islands for example, a complete shifting from flow-through farms into RAS took place after 2000 (Bergheim et al. 2008; 2009). Likewise, over the last decade, there has also been increased interest in the United States for establishing land-based, closed- containment systems (i.e., RAS) to produce salmonid species (Burr et al. 2012). The aforementioned increase has been coupled with an increase of smolts' marketable size range from 50 to 70 g (achieved on traditional flow-through hatchery systems) to 140–170 g achieved in RAS hatcheries (Joensen 2008). For this reason several studies (Davidson et al. 2014; Summerfelt et al. 2013) have subsequently addressed the possibility of developing complete salmon RAS production up to final marketable size. Today, countries producing final marketable size of Atlantic salmon through the use of RAS technology are Canada; China; Denmark; France; Poland and; US (Davidson et al. 2015).

In regard to salmon consumption worldwide, it is now three times higher than it was during 1980. Its success coincides with the rise of both supermarkets' and consumers' interest for a healthy eating life style and salmon products' attributes and format offers (i.e. high fillet yield, fresh, sushi, cured, canned, ready-meals or frozen) (Forster 2010; Asche and Bjørndal 2011; Seafish 2011). The farming industry has converted salmon from a luxury product to a global commodity which is now an affordable staple seafood product for consumers in the industrialized world (Pelletier and Tyedmers 2007; Forster 2010). Nevertheless, many food products face problems when they are put on the market because they do not meet any perceived need for consumers (Asche and Bjørndal 2011). Poor marketing approach and off flavor problems in harvested fish from RAS were reviewed by Badiola et al. 2012). The tendency for the development of off-flavor compounds in the filets of fish cultured within RAS (particularly salmonids) have been widely studied (Schrader et al. 2005; Schrader et al. 2010; Schrader and Summerfelt 2010; Houle et al. 2011; Petersen et al. 2011; Burr et al. 2012). Certain off-flavor compounds can impart a “musty” or “earthy” flavor to the filet which negatively impacts product quality and can result in significant economic consequences (Engle et al. 1995; Tucker 2000). Therefore meeting the consumers demand is a key factor for the viability of any new product to be developed.

Sensory satisfaction is the strongest determinant for fish consumption intention and purchase demand (Verbeke and Vackier 2005). Though consumers may have strong opinions, they usually find it difficult to explain in detail why they prefer one product to another, and the results may be difficult to interpret. Thus, descriptive sensory tests present a sophisticated tool (Lawless and Heymann 1998) and are valuable for product quality analysis, comparison of product prototypes, sensory mapping, and product matching (Gacula 1997). The flesh quality of wild and/or farmed salmon has widely been a subject of sensorial research (Waagbø et al. 1993; Bjerkeng et al. 1997; Einen and Thomassen 1998); some authors have reported quality differences when eating wild and farmed salmon (Skrede and Storebakken 1986; Sylvia et al. 1995), whereas others have found no difference (Higgs et al. 1989). Few of the studies included salmon consumers tests (Sylvia et al. 1995; Farmer et al. 2000).

The current knowledge of land-based salmon production is considerable; development and growth have been studied on RAS (Bergheim et al. 2008; Bergheim et al. 2009; Dalsgaard et al. 2013); fish welfare and performance (Kolarevic and Terjesen 2011), physiological tolerances (Handeland et al. 2004), muscle composition, freshness, texture and colour (Einen and Thomassen 1998), fillet quality and off-flavours (Guttman and Vanrijn 2008; Burr et al. 2012; Davidson et al. 2014), organoleptic (Sylvia et al. 1995), culture engineering (Summerfelt et al. 2013) or economics (Asche and Bjørndal 2011) have been studied on Atlantic salmon from Northern Europe. However, the knowledge on this species (*Salmo salar*) production is still negligible within Southern European countries. Any biological information regarding growth, product quality or economics can be used for predictive models that contribute to develop aquaculture activities at the local level. Such information, in the context of Atlantic salmon, would help to further the experimental RAS culture of this species.

The Basque region is located in the north of Spain. Albeit not aquaculture, fisheries has been for decades the most important income of the primary sector of the region (Zallo and Ayuso 2009). However, most of the exploited fishing areas and commercial species have already reached their maximum potential. Reductions in the commercial landings reflect dramatic changes in the populations of some of the most popular local fish species. During recent years, the policy makers have decided to promote RAS aquaculture with the aim of creating a sustainable and complementary economic activity to the fishing and seafood sectors operating in the region (EJ-GV 2008; 2014). This study prioritized Atlantic salmon (*Salmo salar*) to boost local aquaculture due to native presence of this species in the region (Alvarez et al. 2014) and the clear interest of both local seafood industry and consumers. However, to date no studies on salmon or RAS fish production have been developed in the region.

Therefore, the main objectives of the present study were to: provide new information on the experimental culture of land-based growth of Atlantic salmon (*Salmo salar*) to marketable size in Northern Spain, analyse the effect of temperature on growth, study consumers’ acceptance of the final product, and compare this acceptance between experimentally reared and commercial reared salmon flesh.

2. Materials and methods

The experimental design followed in this study (i.e. location, RAS description, procedures for the water sampling, biological sampling and final product´s analytical composition and sensory evaluation and statistical methods) has been already described in the “Experimental design” section. Target species and study-specific materials and working procedures are presented below.

Individuals rearing

One thousand five hundred Atlantic salmon individuals were obtained from the Marine Institute (County Mayo, Ireland) and transported to the aquaculture land-based research laboratory of AZTI (Getaria, Spain), where several experiments were undertaken. The present study lasted from 30th April 2013 to 9th September 2014. The fish were transported by well-conditioned lorry (i.e. oxygen levels and water temperature were continuously measured during the transport). Thereafter, they were put into 3 equal polycarbonate tanks for acclimation purposes, with running seawater at 10.5 ± 1.05 º C and 31.9 ± 0.61 ‰ during 30 days. The initial mean length and weight (±SD) were 181.1 ± 10.50 mm and 57.67 ± 10.23 g, respectively. In early June 2013, about 250 salmon individuals per tank (9.18 kg m^{-3}) were randomly transferred to another 3 equal tanks which were previously set at seawater's natural thermal range. The experiment´s chronology was set according to Fig. 2.1. It was divided in 3 distinct time periods to accomplish a gradual biological coupling of individuals to the thermal conditions of the region. The first period, called acclimatizing period, lasted one month (from the end of April to beginning June 2013). In the second period, from June 2013 to May 2014, both units worked as partial reuse RAS at a constant 14ºC. Finally, prior to the second summer (i.e. June 2014), both units were divided in two different thermal groups with 3 replicates consisting of: a control RAS unit (i.e. control unit; C1, C2 and C3) with a maximum thermal set of 14 ± 1.5ºC and a natural RAS unit (i.e. natural unit; N1, N2 and N3) with a maximum thermal set of 19 ± 1.5ºC. This period was extended until end of September 2014.

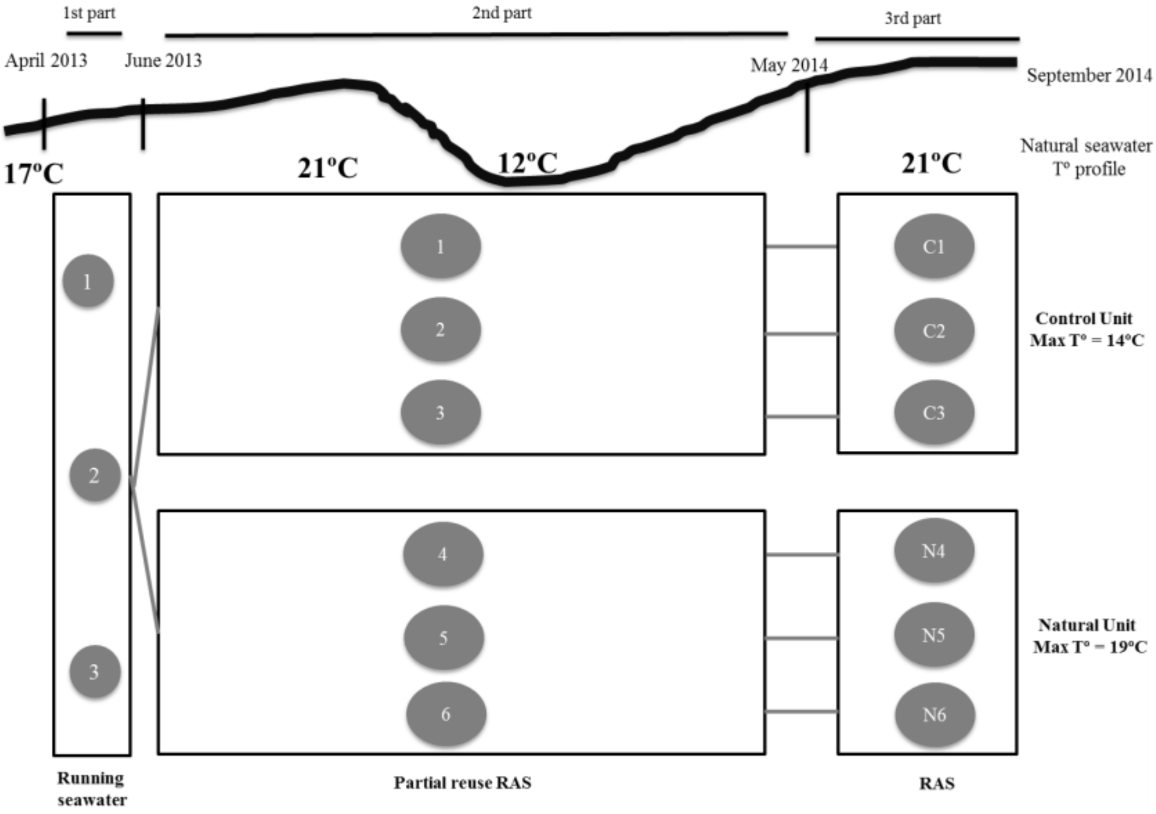


Fig. 2.1. Experiment's chronology and methodology.

A 16 h light: 8 h dark photoperiod was employed. Identical food conditions were offered to individuals in each of the 6 tanks; this was arranged by organising the feeding schedule based on a day's degree scale (i.e. days after containment per temperature). Fish individuals were fed 3 times a day (8:00, 12:00 and 16:00) and they were not treated with any antibiotic solution. Daily food ration was adjusted to 0.8% of fish biomass per tank and pellet size (Spirit, Skretting S.A., Burgos, Spain) ranging from 3 to 9 mm during the whole experiment. The uneaten feed was collected on a daily basis. All individuals were visually checked on a daily basis to mitigate mortality events.

Immediately after arrival, 30 fish individuals were measured and weighed to the nearest 0.01 cm and 0.1 g, using an ictiometer and a precision balance (Mettler Toledo), respectively. For subsequent growth and condition determinations, 24 length (SL) and weight measurement were taken from both treatment groups every 2-3 months after-containment. For this purpose, salmon individuals were starved for 24 h prior to sampling and then anesthetized with 30 mg L^{-1} clove oil in seawater. Body weight (W) was calculated, to the nearest 0.1 g, on a microbalance (Mettler Toledo). Instantaneous rate of growth (*G*) and the relative rate of growth (*K*) were estimated as follows (Ricker 1975):

$$W_t = W_0 \cdot e^{G \cdot t} \quad or \quad L_t = L_0 \cdot e^{G \cdot t}, \;\; and \quad K = e^G - 1$$

where W_0 and L_0 are the initial DW (in g) and the SL (in cm); and W_t and L_t are DW and SL, at time *t* (in days), respectively. For each case, the daily specific growth rate was defined by *K* x100 (%) and at each treatment group, the SL-at-age data were fitted by linear regression to test the suitability of the exponential model. Individual mortality was registered on a daily basis.

Sensory evaluation

Two fresh salmon products from two different production origins were evaluated: locally grown (i.e. from the present study and not commercial) and Danish farmed salmon (i.e. 5 individuals produced by a RAS company located in *Hirtshals (Denmark)* were bought in the local market for the evaluation). The consumers group included 63.3 % females and 36.7 % males. The average age was 36 years; divided over age ranges: 18-30 years (13.3 %); 31-40 (40 %); 41-50 years (33.3 %) and; 51-60 years (13.3 %). 87 % of the consumers were regular salmon fish consumers; divided over consumption frequency: from time to time (13.3 %); once per month (56.7 %); once per week (26.7 %); and more than once per week (3.3 %).

3. Results

Individuals rearing

Water quality parameters results are shown in Fig. 2.2. All parameters (i.e. dissolved oxygen, pH, ammonia, nitrite, nitrate, temperature, bromide, chloride and salinity) were successfully maintained within the security levels for correct survival and growth. Temperature varied between 9.1-15.6 ºC and 9.5-20.5 ºC in control and natural units, respectively. In terms of mortality, no significant differences (ANCOVA, $P > 0.05$) were observed between both RAS units. At the end of the study the cumulative survival rate ranged from 85% to 75% between the natural and control units, respectively (Fig. 2.3).

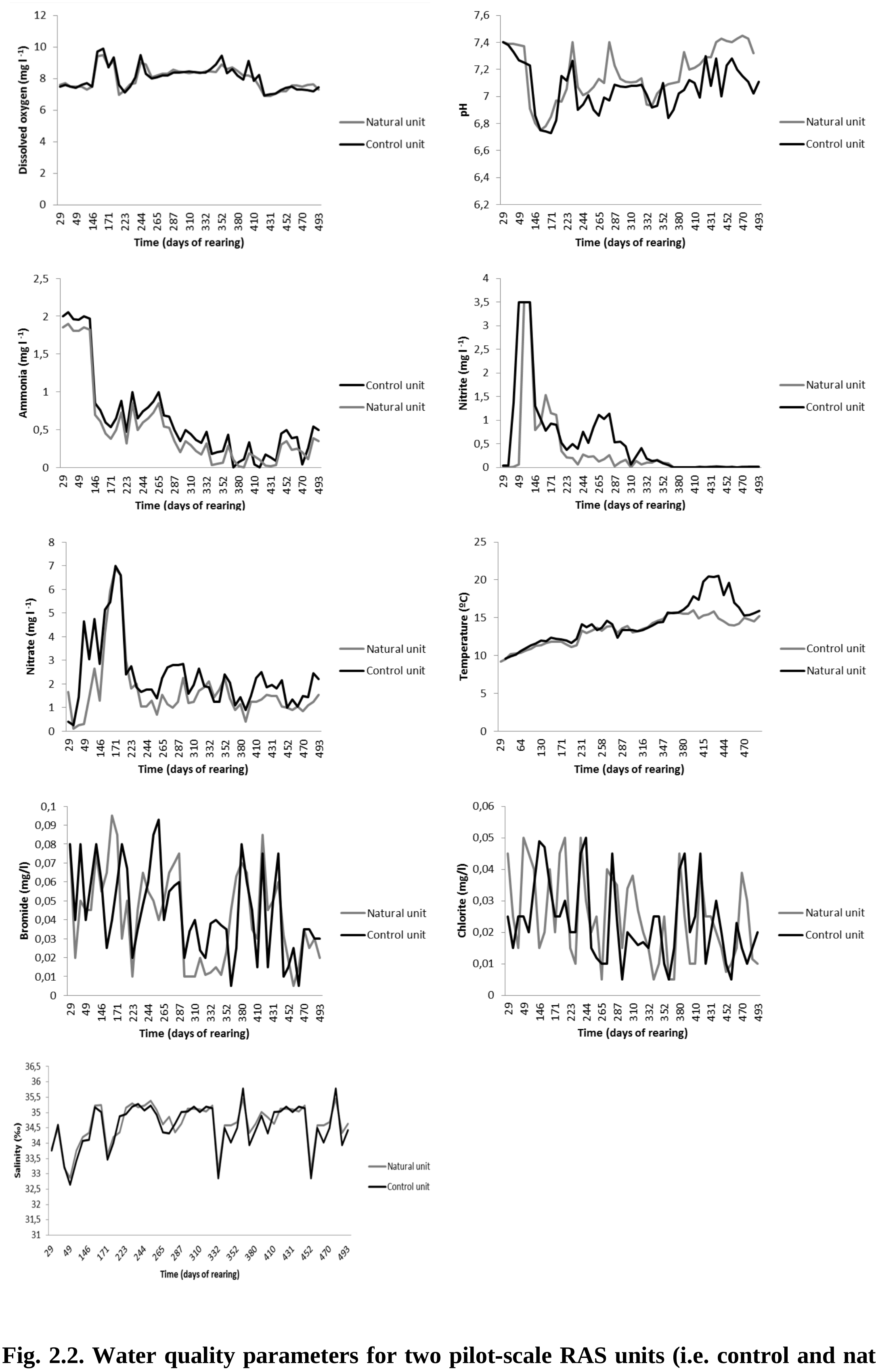


Fig. 2.2. Water quality parameters for two pilot-scale RAS units (i.e. control and natural units) along the experiment.

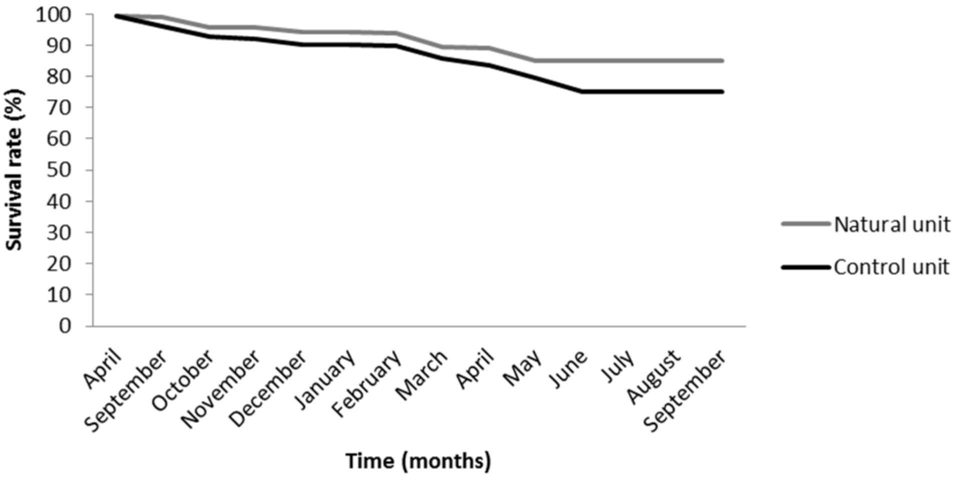


Fig. 2.3 Survivorship curve (lines) taking temperature as a factor. The lines represent the observed surviving rate of salmon (*Atlantic salmon*) individuals.

The exponential equation provided a good fit for growth in length-at-age, explaining between 91 and 93% of the variability in L_t for the control and natural unit, respectively. For each module, the parameters of growth equations are listed in Table 2.1. Daily specific growth rates were affected by sampling time showing significant differences (ANOVA, $P < 0.05$) from the beginning of winter and the end of spring; natural unit's individuals suffered a halt (SGR decreased from 1 to below 0.5% day^{-1}) after the first summer (Fig. 2.4). Nevertheless, from late spring 2014 onwards, the natural unit's individuals showed a compensatory recovery and kept growing, but without significant differences from the control unit's individuals (ANCOVA $P > 0.05$). At the same time, the control unit's individuals stopped growing, suffering a decrease in the average growth (Fig. 2.4 - growth graphic, day 420) and maintaining below the natural unit until the end of the experiment. Both condition and hepatosomatic indexes were also affected (ANOVA, $P < 0.05$) by sampling time. While time (i.e. season) had the most significant impact on daily specific growth rates, at the end of the study no significant differences (ANOVA, $P > 0.05$) were detected between individuals from different regimes. Food Conversion Ratio (FCR) was 1.51 ± 0.30 and 1.23 ± 0.47 at control and natural units, respectively. No significant differences in FCR were found ($P > 0.05$) at the end of the experiment.

Table 2.1. Growth values: (A) Estimated parameters of the length-at-age exponential growth equation for each thermal experiment. Equations are given in text. (B) Instantaneous (G), relative (K) and daily specific (%) growth rates, of weight, calculated from the equations reported by Ricker (1976). Statistical significance (*) was set up at ($p < 0,05$) (pairwise comparison).

Control unit (14 ± 1.5ºC)					Natural unit (max. 19 ± 1.5ºC)				
W0	**G**	**K %**	**Nº observations**	**R^2**	**W0**	**G**	**K%**	**Nº observations**	**R^2**
57.66	0.0062	0.625	231	0.95	57.66	0.0098	0.990	231	0.94
L0	**G**	**K%**	**Nº observations**	**R^2**	**L0**	**G**	**K%**	**Nº observations**	**R^2**
18.11	0.0020	0.201	231	0.91	18.71	0.002	0.201	231	0.93

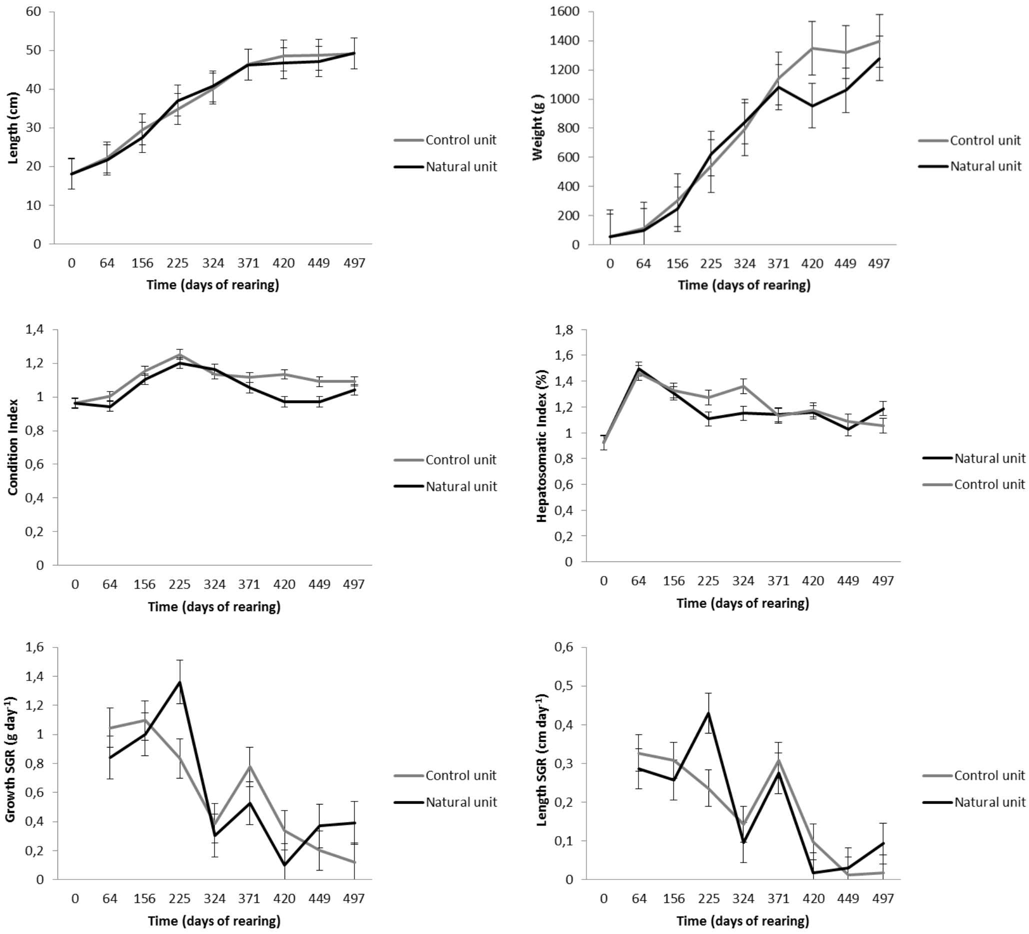
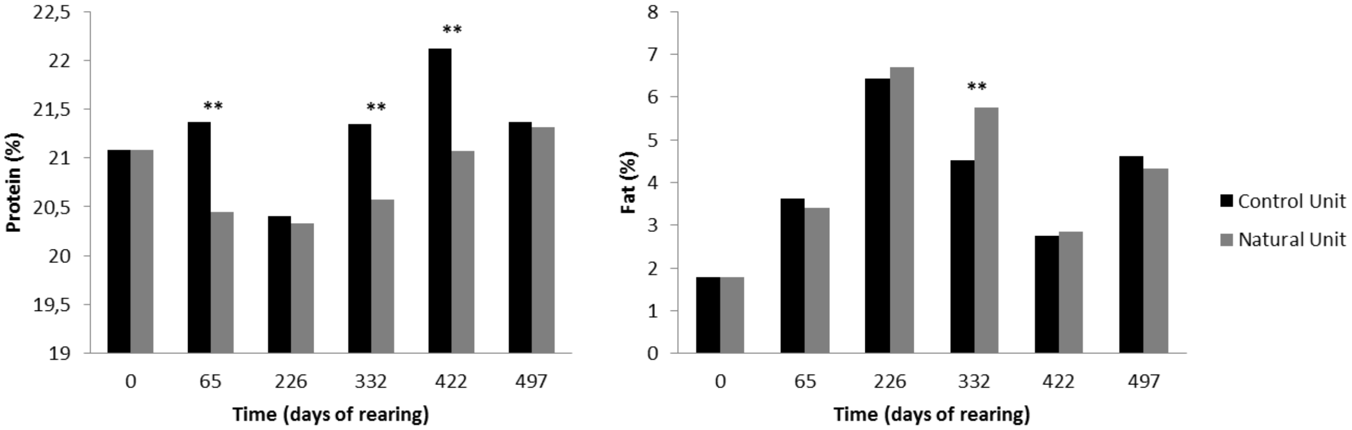


Figure 2.4 Length (cm), weight (g), Condition index, Hepatosomatic index (%), Specific Growth Rate (% day^{-1}), and Length Specific Growth Rate (cm day^{-1}) for two pilot-scale RAS units (i.e. control and natural units) along the experiment.

Analytical composition

The results of the proximate analytical composition (presented as protein and fat) are shown in Fig. 2.5. Significat differences ($P < 0.05$) were found in terms of protein content in two of the samplings after the winter period although no differences ($P > 0.05$) were found at the end of the experiment. In terms of fat content, significant differences ($P < 0.05$) were found in the sampling made just after the winter period.



Sensory analysis

Results from the sensory analysis made by the consumers are shown in Table 2.2. Significant statistical differences ($p > 0.05$) between both of sample groups (i.e. locally grown and commercial salmon) were not found for any of the studied characteristics (i.e. aspect, smell, flavor and texture). Locally grown individuals and both origins fillets are shown in Fig. 2.6.

Table 2.2. Results from the sensory evaluation made by the consumers.

	Locally grown salmon		Commercial salmon		t Student	
	Mean[1]	SD[2]	Mean	SD	t	*p*-value[3]
Aspect	6.200	± 1.400	6.567	± 1.331	1.040	0.303 ns
Smell	6.633	± 1.129	6.700	± 1.442	0,199	0.843 ns
Flavor	6.867	± 1.196	6.800	± 1.960	-0.176	0.861 ns
Texture	6.667	± 1.213	6.767	± 1.478	0.286	0.776 ns
Global impression	6.733	±1.081	6.833	±1.487	0.298	0.767 ns

[1] mean from each sample
[2] standard deviation
[3] ns no significant; * significant at 5 %; ** significant at 1 %; *** significant at 0.1 %

Figure 2.6 Locally grown salmon individuals (A, B, C); locally grown and commercial salmon fillets (D), and locally grown and commercial salmon samples ready to cook for the tasting panelists (E).

Both of the salmon products were accepted equally (Fig. 2.7). Results did not show any significant differences ($P > 0.05$) at sensorial level between the samples. Global impression was around 7 points (in a scale from 1 to 9) for both locally grown and commercial salmon (6.733 and 6.833, respectively) not presenting significant differences ($p > 0.05$). The hedonic study revealed that the consumers evaluated both samples as “I like it quite a lot”. The reported perceptions by the consumers to describe both locally grown and commercial salmon are shown in Table 2.3. In general terms, locally grown salmon’s characteristics were described as good aspect, particular salmon smell and flavour with an adequate intensity whereas the commercial salmon was described as drier but stronger flavor. In contrast, fillets color was described as slightly pale and less succulent texture in comparison with the commercial salmon.

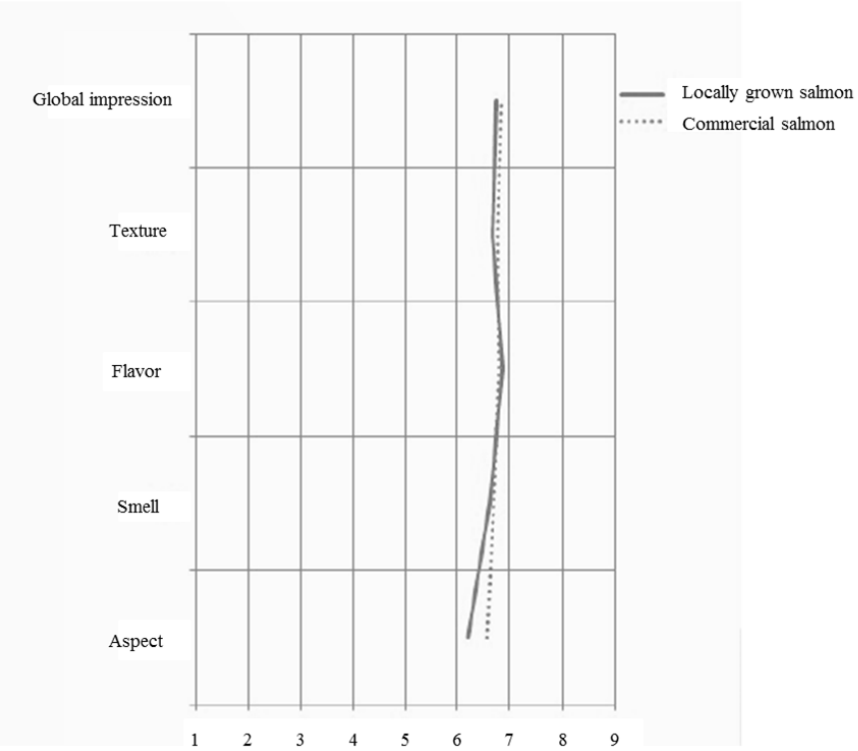


Fig. 2.7. Consumers' acceptance level of studied characteristics (i.e. aspect, smell, flavor, texture and global impression) for both locally grown and commercial salmon.

Table 2.3. Consumer perceptions while describing locally grown and commercial salmon.

Locally grown salmon	Commercial salmon
More succulent	Too dry, thick and oily
Whitish meat, milky smell	Very pale color, more succulent
Harsh texture, leathery	Great taste and texture
Very pale color	Pinker color, oilier
Very pale meat, not much smell and flavour	Lightly rusted smell, rubbery
Slightly dry	Drier, rubbery texture
Rubbery texture	Drier, unknown flavor
Salmon taste and smell, less succulent	Better color, more orange-colored
Pale color with a breakable texture	Little strong flavor and a bit dry

Results from the assessment about consumers' fish purchasing intention are shown in Fig. 2.8. 80 % of the consumers would definitely buy the experimental locally grown product tested. The other 20 % were divided between consumers that did not know if they would buy or not (10 %) and consumers that would more likely would not buy it (i.e. 3.3 % probably not and 6.7 % definitely not). Likewise, 70 % of the consumers would definitely purchase commercial salmon while the rest 30 % was divided, 20 % were in doubt and 10 % would definitely not buy it.

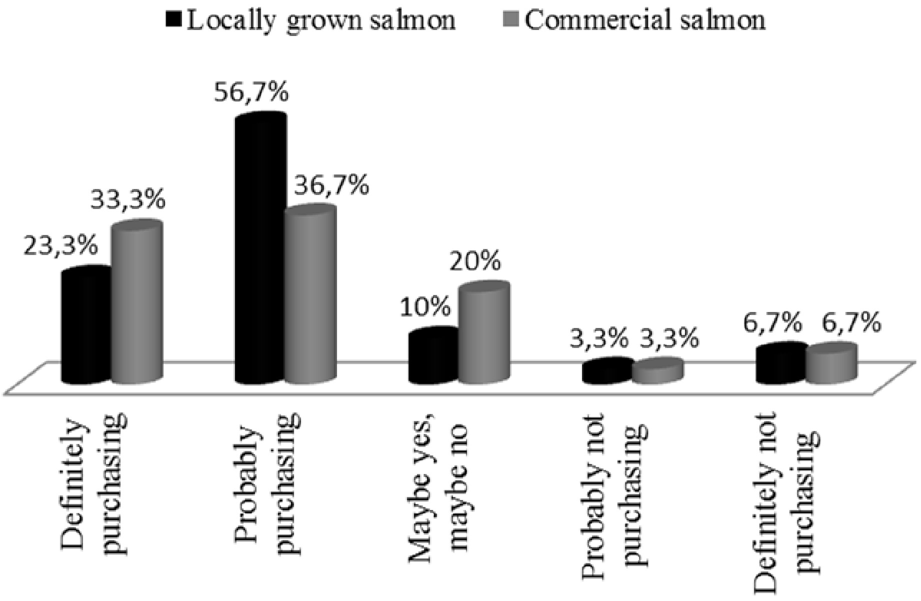


Fig. 2.8 Locally grown and commercial salmon consumers' purchasing intention.

4. Discussion

This is the first study on land based on-growing of Atlantic salmon to marketable size in the North of Spain. Normally, at approximately 60–90 g, Atlantic salmon fingerlings undergo a physiological transformation (i.e. smoltification) and can adapt to seawater being transferred to marine net pens until harvest. Thus, grow-out mainly takes place in net cages at sea, while smolts traditionally have been produced in freshwater land-based systems (i.e. RAS and flow-through) and in cages and lakes (Bergheim et al. 2009; Terjesen et al. 2008). Currently, the size of transfer has been varying from 30 – 50 to 140-170 g (Bergheim et al. 2009) and future plans are to grow them until 1 kg on land (Dalsgaard et al. 2013) using freshwater.

Atlantic salmon grow faster at 12 ‰ due to less energy demand for osmoregulation (Warrer-Hansen 2015). Thus, some of the mortality that occurred during the acclimation period could be due to stress post-transportation (Iversen et al. 1998; Iversen et al. 2005), and the fact that some of the individuals were not prepared for their introduction to 33-35 ‰ salinity waters. This cause it is not yet verified, but should be considered carefully in RAS where the stocking density is higher than cages and individual mortalities can lead to a chain reaction die offs.

In general, both pilot-scale RAS units (i.e. natural and control units) showed acceptable water quality and mortality rates, albeit some differences in growth performance were found. Both biofilters performed adequately, maintaining appropriate and lower concentrations than the established limited threshold of ammonia, nitrite and nitrate along the experiment (Kolarevic and Terjesen 2011). This was achieved by controlling feeding and fish stocking density, as well as maintaining oxygen concentration above saturation levels. The possibility that certain levels of nitrate could be chronically toxic and/or act as an endocrine disruptor to some RAS-produced finfish is still not clear (Suzuki et al. 2003; Klas et al. 2006; van Rijn et al. 2006). Nevertheless, a recent study has demonstrated that post-smolt Atlantic salmon can be safely cultured in RAS at nitrate levels up to 100 mg l^{-1} (Davidson et al. 2015), levels that are above the maximum concentrations achieved in the present study in both natural and control units (6.9 and 7 mg l^{-1}, respectively). Increasing the threshold limit for nitrate concentrations would reduce the use of the make-up water. This reduces the discharge flow that is required for treatment and increases heat retention of the water, saving energy and costs related to pumping and heat retention. Furthermore, the observed constant reduction on nitrate concentration in the present study as the experiment progressed corroborates the adoption of a partial water reuse strategy as a method to achieve a gradual but successful bio coupling of salmon individual to the conditions of the new environment. Albeit, this referred a slow process, the conditions are suggested to be considered a minimum requirement to secure salmon acclimation procedures within the case study region. Likewise, the aforementioned procedure is initially recommended for experimental scenarios but for commercial operation further investigations should be developed in order to reduce the time elapsed.

Regarding growth performance, a compensatory growth (Jobling 1994) was observed during the time of rearing within the natural pilot-scale RAS unit. Experiments with Atlantic cod (Luczkovich and Stellwag 1993; Imsland et al. 2005b; Badiola et al. 2016) and halibut (Larsen et al. 2010) have showed that an early environmental manipulation (i.e. temperature) towards colder water could lead to a long-term effects, stimulating benefits in commercial aquaculture (e.g. faster growth in a given timeframe). In average, natural pilot-scale RAS unit experienced higher temperatures. The difference was greater in the second summer period (i.e. from late June to beginning of September) when differences oscillated from 4.5 to 5.6ºC between the day 415 and the day 452. It was in this period when individuals from the natural pilot-scale RAS unit experienced a recovery, increasing weight and overtaking individuals from the control pilot-scale RAS unit. Fish and other animals are able to adapt to feast-and-famine conditions by showing marked growth spurts when environmental conditions (i.e. temperature) are improved after a period of less favorable conditions (Jobling 1994). Moreover, fish that are in the poorest physical condition show the greatest response, displaying immediate mortality or the most rapid rates of weight gain when adequate rearing conditions are restored (Jobling 1994). Thus, the individuals from the natural pilot-scale RAS unit were not affected by higher temperatures, neither was their appetite, mortality, or growth. Additionally, the analytical composition showed that both temperature regimes ended with similar fat and protein contents. Few publications have been published about salmon fillet’s composition reared in RAS. About the fat content, obtained values were similar, in absolute terms, than those concluded by Burr et al. (2012). Conversely, the protein content was higher than the ones obtained by Bjerkeng et al. (1997) in sea-cages.

The combination of characteristics such as wholesomeness, integrity and freshness define the quality of fish (Martin 1988) and descriptive analysis is undoubtedly one of the most valuable sensory tools to provide detailed information on product quality and specific properties (Murray et al. 2001) from the consumers. The use of such procedure has been widely studied in a wide range of food and beverages in the food industry: wine (Heymann and Noble 1987; Francis et al. 1992); milk (Hough and Sánchez 1998; Torres-Penaranda and Reitmeier 2001; Chapman et al. 2001) and; fruit (Shamaila et al. 1992), for example. In the present study, two different origin farmed salmon products were tasted by the consumers, resulting in positive feedback. In fact, the use of hedonic scaling methods to assess the acceptability of the salmon farmed over one year and five months in RAS, shows that, in general, the locally grown salmon were at least as acceptable as the commercial salmon. Consumers were not able to discriminate between samples and the resulting scores were almost equal across the board with respect to studied attributes (i.e. aspect, smell, flavor and texture) and global impression; all these results indicate consumers´ acceptability of the products tested.

One of the issues that concerns fish farmers is the “musty” or “earthy” flavor that tends to develop in the filets or fish reared in RAS, particularly in salmonids (Schrader et al. 2005; Houle et al. 2011; Petersen et al. 2011; Burr et al. 2012) and the potential economic losses that it may provoke (Engle et al. 1995; Tucker 2000). The persistent off-flavor compounds are naturally caused by organic chemicals geosmin and 2-methylisoborneol (MIB) (Persson 1980; Schrader et al. 2005; Guttman and Vanrijn 2008; Schrader and Summerfelt 2010; Houle et al. 2011). These are secondary metabolic products of certain species of cyanobacteria and actinomycete bacteria (Slater and Blok 1983), primary producers of the persistent off-flavor compounds in RAS (Guttman and Vanrijn 2008; Schrader and Summerfelt 2010). These off-flavor compounds are likely to be persistent to the cooking process of the fish (Farmer et al. 2000); however no unpleasant smells and/or flavor were reported by the consumers of the present study for any of the tasted samples. Likewise, the locally grown experimental salmon individuals resulted in a better scored product by the consumers than the commercial product (6.876 and 6.800, respectively), although statistically there was not significant difference.

The aroma and flavor of farmed Atlantic salmon have been shown to become less intense as maturation progresses (Aksnes et al. 1986; Blokhus 1986). These changes occur after the skin color has changed from silver to brown and at the same time as the underjaw changes shape (Aksnes et al. 1986). However, the salmon used in this study did not show any visible signs of maturation in terms of altered shape or color of the whole fish and the color resulted to be pale to very pale and no negative comments were reported.

Freshness is one of the most important quality criteria for buyers of Atlantic salmon and starvation is performed routinely prior to slaughter to ensure that the fish have an empty gut to improve the quality of such criteria (Blokhus 1986). Short and long-term starvation periods have been studied (Blokhus 1986; Johansson and Kiessling 1991) resulting in different effects on raw and cooked fish flesh. Moreover, the latest study (Einen and Thomassen 1998) concluded that starvation (i.e. 0 to 86 days) prior to slaughter was a weak tool for changing fillets quality in Atlantic salmon (i.e. raw and cooked) due to the fact that only small changes were found in freshness, texture and color characteristics. Thus, in the present contribution, 24 hours of starvation was applied and no significant differences were found with the commercial salmon in any of the studied characteristics. Furthermore, freezing and frozen storage of fish has also been studied due to the possible changes in the muscle proteins (Shewfelt 1981; Mackie 1993). The decreases in juicy and moist appearance, and in separation, also observed on freezing, are likely to be a consequence of a reduction in water holding capacity arising from protein damage. Herein, fishy odors can develop when fish is stored for long periods or under inadequate conditions due to trimethylamine or dimethylamine, formed in marine fish from trimethylamine oxide by the action of microbial or endogenous enzymes, respectively (Lindsay 1988). In the present study it was assumed that commercial salmon were harvested and frozen in storage for longer than the locally grown salmon which was harvested 24 hours prior to evaluation. Nevertheless, there was no evidence of any relationship between frozen storage time and flavor or odor attributes as no significant differences were found between both samples and thus no effects of such procedure (i.e. frozen storage) were detected.

Different studies have verified the health benefits of seafood consumption (De Deckere et al. 1998; Marckmann and Grønbaek 1999), concluding that the intake of at least 1-2 times of fish per week has a positive effect on health. Moreover, the consumption has been positively correlated to its easy and convenient access (Olsen et al. 2007), while fish price has been found to be one of the main barriers (Verbeke and Vackier 2005). In Europe, the average per capita consumption of fish is estimated around 24.9 kg per year (6 kg more than in the rest of the world) (COM 2016) while in Spain is 26.4 kg per year (MAGRAMA 2015). In the Basque region these values stay behind the average which turns to be 36.9 kg per year (Mercasa 2014). Additionally, fish represents the second most important food in terms of level of expenditure behind the meat and prior to fresh fruits and vegetables (Mercasa 2004). In the present contribution, purchasing intention of the locally grown salmon resulted to be more likely than the commercial salmon (i.e. 80 and 70% of the consumers, respectively), remarking the positive acceptance that consumers would show towards a possible marketable product from the region. Moreover, consumers that were in doubt were likely influenced by other factors such as price and seasonality of the product. In this way, future research efforts should be directed towards marketing strategies and consumers' education, fulfilling the knowledge gaps and economic requirements of this potential fish species for the market of the case-study region.

5. Conclusions

The results of the present study constitute the first available data on growth performance and organoleptic of Atlantic salmon (*Salmo salar*) cultured within land-based scenarios of Northern Spain. The experimental results of the present aquaculture study are useful to biologists, farm manager's or policy makers to understand biological feasibility, infer productivity or recommend management practices for further production activities. As reported by the consumers of the present study, Atlantic salmon refers an attractive and suitable candidate species to be grown locally.

Contribution 3

Energy use in Recirculating Aquaculture Systems: a review.

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Badiola, M., Hundley, P., Basurko, O.C., Piedrahita, R., Mendiola, D. 2017. Energy use in Recirculating Aquaculture Systems: a review. *Environmental Science and Technology* (under revision).

Summary

Recirculating aquaculture systems (RASs) are intensive fish production systems, with reduced use of water and land. Nevertheless, their high energy requirement is a drawback, which increases both operational costs and the potential impacts created by the use of fossil fuels. Energy use in RAS has been studied indirectly and/or mentioned in several publications. Nevertheless, its importance and impacts have not been studied. In aiming to achieve economic and environmentally sustainable production a compromise has to be found between water use, waste discharge, energy consumption and productivity. The current review discusses published studies about energy use and RAS designs efficiencies. Moreover, with the aim of making an industry base study a questionnaire about the energy use in commercial scale RAS was conducted. The design of more efficient and less energy dependent RAS is presented, including optimized unit processes, system integration and equipment selection. The main conclusions are: fossil based fuels are less cost-effective than renewable energies; energy is of little concern for the majority of the industry, and renewable energies are of potential use in RAS.

1. Introduction

Animal farming, including fish farming, may cause significant environmental problems such as resource depletion as well as contributing to climate change (Winther et al. 2009; Sonesson et al. 2010; Lesschen et al. 2011; Nijdam et al. 2012). Intensification of farming practices (Steinfeld and Wassenaar 2007) together with the steady increase in the demand for fish (FAO 2014) has pushed the aquaculture industry to look for acceptable practices from environmental, societal and economic perspectives. In aquaculture, water and energy are two of the main resources to be considered (d'Orbcastel et al. 2009b). They are indeed the baseline for industry development (COM 2002; COM 2009; NOAA 2011a). Consequently, an improvement in water management will aid aquaculture's progress (Dumont et al. 2012), which has slowed down recently for some forms of fish farming (e.g. flow-through systems) (Naylor et al. 2000; Buschmann et al. 2006).

Compared to other forms of aquaculture production, recirculating aquaculture systems (RASs) decrease potential environmental impacts such as eutrophication as well as water dependence (Verdegem et al. 2006; d'Orbcastel et al. 2009b; Eding et al. 2009) aiding waste management (i.e. reduced waste volumes) and boost nutrient recycling (Piedrahita 2003). RAS are intensive fish production systems, with reduced water and land use. Nevertheless, their high energy requirement is a challenge which increases operational costs (Aubin et al. 2006; Colt et al. 2008; d'Orbcastel et al. 2009b). Thus, on-farm electricity consumption affects both environmental impacts and economic costs (i.e. operational costs) of a RAS (Badiola et al. 2017), jeopardizing the farms' sustainability. Currently, there is interest in using renewable energy sources or waste heat from other industries as part of the solution to decrease environmental impacts due to the use of fossil fuels. Nevertheless, the energy source to be employed in a farm will be dictated by the system's location and accessibility to the energy sources. The location of aquaculture operations, sometimes in remote areas, may make it easier to use renewable energy than in other industries.

In 2013, the food sector was a major consumer of energy, accounting for 26% of the European Union's final energy consumption (Monforti-Ferrario et al. 2015). Indeed, agriculture and livestock were responsible for 33.4 % of the energy embedded in the food consumed, representing the largest contributing sector. Moreover, the energy consumed in the fishery sector (including aquaculture) was equivalent to almost 5 % (i.e. 45 Petajoule) of the direct energy consumed in the agriculture sector. Clearly, energy plays a vital role in industrial and technological developments around the world (Dincer 1999; Midilli et al. 2005a; 2005b).

A possible solution to decrease energy usage and increase production efficiencies may be creating energy efficient production systems. This is recognized as a cost-effective way of addressing the wide-ranging problems associated with: the changing global energy scene (i.e. reducing dependence on fossil fuels while increasing the use of renewable sources); mitigation of greenhouse gas emissions from industry (Worrell et al. 2009); and industry's economic competitiveness promoting cost savings (Worrell et al. 2003). In fact, tracking sector-wide energy efficiency trends has grown in importance (Ang et al. 2010) due to its direct relationship with the improvement of industrial process productivity (i.e. lower capital and operating costs, increased yields, and reductions in resource and energy use) (Kelly et al. 1989; Boyd and Pang 2000).

In RAS, as in other forms of aquaculture, operating costs should dictate the most efficient design. Little has been published about the energy use and energy efficiency in RAS. The few examples include the work done by Colt et al. 2008; d'Orbcastel et al. 2009b; Buck 2012; Ioakeimidis et al. 2013.

The main objective of this paper is to provide an in-depth analysis of the energy demand of RAS aquaculture. Current trends in energy use, energy sources and energy efficiency in the sector are analyzed based on an extensive literature review (including over 200 publications and 58 books) and a survey of stakeholders' points of view regarding energy-related challenges. Finally, results are used to propose optimized RAS unit processes, engineered system integration, and equipment selections as guidance for designing RAS farms. Alternative design solutions for each system, subsystem, and component are presented as well.

The mentioned extensive literature review has concluded that there is not many information regarding to the energy use in RAS. In that way, some of the citations along the manuscript are old. The authors of the paper agree that some of the facts may have changed positively with the time although there are not evidences to proof such improvement.

2. Energy use in RAS:

Energy use and its associated cost and environmental impact depend on the source and quantity of energy used, location, design and management. The following section discusses the implication of design on efficiency and energy use. It also provides an overview on the most frequent equipment and processes used in RAS, different energy sources, and energy consumption values for production of different species.

2.1. Efficient design and energy use by equipment and/or processes:

In RAS water is re-used after undergoing different treatments (i.e. water treatment loop); the remainder, after being treated, is discharged into an appropriate water body (e.g. the sea, a lake, a river). Hence, an equal amount of clean water from an external water body (e.g. the sea, a river, a municipal water source) is pumped into the RAS system to maintain a constant volume of water (Rosenthal et al. 1986). The water treatment loop is formed by different unit operations. Some require energy (e.g. pumps); others (e.g. biofilter) influence the energy consumption due to their design and/or management (e.g. equipment height determines pumping head and energy needed), despite not needing energy directly to power the equipment. Each RAS is different and the technology to be used in the water treatment loop may differ between systems. Thus, the operations requiring energy use in RAS and the overall energy requirement will be determined by engineering and operational criteria, such as: water circulation including pumping of the incoming water and of water through the treatment loop; heating/cooling of water; oxygenation; filtration and/or removal of solids and nitrogen compounds; stripping of CO_2 and; disinfection, and ozonation.

Table 3.1 presents a summary of the main advantages and disadvantages of the usual technology used in RAS. The comparison has only focused on their efficiency. Some examples from the literature on their energy requirements and costs are also presented.

Table 3.1. Main advantages and disadvantages of some common technologies employed in a water treatment loop

	Equipment	Advantage	Disadvantage	Energy consumption/costs	References
Water moving - Pumps	Centrifugal pumps	Effective for moving water at high head	Increased energy consumption		Mayo, 1976; Wheaton 1977
	Airlift pumps	Inexpensive Simple to use Combine several functions Reduce space used	Not sufficient in high-density RAS	Lower energy costs than centrifugal pumps	Blancheton et al 2007; Mamame et al 2010; Barrut et al 2011,2012; d'Orbcastel et al 2009
	Axial flow pumps	Large volumes at modest heads	Low water lift	Lower energy costs than centrifugal pumps	Timmons and Ebeling 2010; Barrut et al 2012
Oxygenation vs.aeration	Oxygenation	Additional safety Maintenance of fully saturated conditions.	Cost-effective in large scale RAS Distribution/maintenance requirements Increased operational costs	Small fraction of the total energy used in large-scale RAS hatchery	Sowerbutts and Forster 1980; Colt et al 2008
	Aeration	Simple to manage Little maintenance required Inexpensive equipment	Limited efficiency May be difficult to measure	Significant fraction of the total energy consumption	d'Orbcasterl et al 2009
Solids filtration/removal	Sedimentation	Proves a suitable process for clarification of lower flow rates (e.g. sludge flow produced by a screen separator)	Insufficient residence time to particle settle out; scouring of settled particles off the bottom; short circuiting of influent water direct to the outflow. Not suitable for clarifying untreated main wastewater flow from a farm.		Cripps and Kelly, 1996; Summerfelt, 1998; Cripps and Bergheim 2000
	Static screens		Capacity limitations,		Makinen et al (1988)

	Rotating microscreens	Suitable where blockage is likely Potential to gently remove particles with minimal damage			Wheaton 1977
	Rotating screens	Backwashing sludge can be reused/applied to farmland	Substantial backwash sludge which requires further thickening/dewatering		Bergheim et al 1998
	Rotating disc screens		Limited capacity in comparison with disc screens		Cripps and Bergheim 2000
Wastewater sludge removal/filtration	Gravity thickening settlers	Concentrated biosolids are land applied, composted or hauled to a landfill		0.763 kWh/unit	Henderson and Bromage1988; Bergheim et al. 1998; Chen et al. 1997, 2002; Brazil and Summerfelt, 2006; Sindilariu et al., 2009; Sharrer et al 2010
	Inclined belt filters	Reduces TAN leaching as rapidly separates biosolids from wastewater	More mechanically complex than geotextile or bag or gravity thickening settlers	24.95 kWh/unit (includes: solids pump; clarified water pump; belt filter; mixing tank mixer; polymer and alum storage mixer; polymer and alum dosing pump)	Ebeling et al., 2006; Summerfelt and Vinci 2008; Sharrer et al 2010
	Geotextile bag filters	Dewatered biosolids suitable for land application, composting, incineration or landfill. Increased TAN leaching as solids are stored in anaerobic conditions	Require the application of a polymer to enhance floc formation	19.15 kWh/unit (includes: permeate pump; polymer and alum storage mixer; polymer and alum dosing pump)	Sharrer et al., 2009; 2010
Nitrogen removal	Rotating biological contactor	Plug-flow pattern increases removal efficiency Low head requirements Passive aeration CO2 removal	High costs; low volumetric efficiency		Miller and Libey, 1985; Brazil 2006

Trickling filters	CO2 removal by degassing Water cooling in summertime; Simplicity of design, construction, operation and management.	Relatively low volumetric removal rates (i.e. large sized biofilters) Risk of clogging Additional solids removal necessary Relatively high pumping cost	Models to predict energy costs but many are variables affecting the energy use	Kamstra et al., 1998; Schnel et al., 2002; Eding et al., 2006; Lyssenko and Wheaton, 2006; Crab et al 2007
Moving bed bioreactors (MBBR)	Low head loss; high specific biofilm surface area; no backwashing needed; low maintenance; small footprint	The efficiency is highly dependent on the media used and working parameters fluctuations (e.g. temperature)		Rusten et al. 1995; Zimmerman et al. 2005; Ødegaard 2006; Rusten et al. 2006; Bjornberg et al. 2009; Pfeiffer and Wills 2011; Qiqi et al. 2012
Downflow microbead filters	Smaller media, increased surface area High hydraulic loadings possible			Greiner and Timmons, 1998; Timmons et al., 2006
Fluidized sand biofilters	High specific surface area Moderate cost	No aeration Narrow water flow range High pumping cost		Miller and Libey, 1985; Timmons and Summerfelt, 1998
Fluidized bed filter using plastic media	High specific surface area per unit volume (reduced hydraulic retention) Reduce energy costs	High head loss, increasing energy requirements		Summerfelt and Cleasby, 1993; Honeyfield and Watten, 1996; Sandu et al 2002;

Water circulation

In RAS, water is usually circulated by pumps to move water to a higher elevation or to increase the overall system pressure for filtration, aeration and degassing. Depending on a system's hydraulics, there are two RAS types: pressurized or high-head systems and low-head systems. The advantage of a pressurized or high-head RAS is the hydraulic link between source and the point of discharge, which is relatively independent of the pipe's geometry. However, a change in flow at one distribution point will influence flow at another point. In such systems centrifugal pumps are used, which efficiency depends on the impeller's design, limiting the size of solids that passes through the pump. In contrast, low-head RAS present the advantage of moving large volumes of water using significantly lower energy, improving the economic returns of investment (Pfeiffer and Wills 2008; Pfeiffer and Riche 2011). In such systems, either the airlift pumps, axial-flow propeller pumps or some combination of the two is used.

The capacity of airlift pumps has been generally thought to be insufficient to provide the water treatment requirements of high-density RAS, while axial flow pumps may be efficient at moving large volumes of water to modest head levels (e.g. 4.6 to 9.2 m) and tolerable to small debris and solids (Timmons and Ebeling 2010). The main disadvantage with airlift pumps is the low water delivery height, which is limited to a maximum of around 0.3 m. In those cases, the energy needed could be reduced by 40% compared to centrifugal pumps (Barrut et al. 2012). The head loss in most RAS is a limiting factor; operating costs can increase 20-40% at 1 m pumping head and over 44-69% at 3 m head (Muir 1978).

Recently, due to the high operational costs of pumping (Dunning et al. 1998; Colt et al. 2008), airlifts are becoming more common (Blancheton et al. 2007; Mamane et al. 2010); they are simple to use and economic under a limited set of operating conditions. Moreover, this equipment can serve for water transport, gas exchange and foam fractionation (Barrut et al. 2012), which may have some advantages when compared to other pumping methods, such as a lower occurrence of breakdowns, a reduction of the need for technical supervision, and a reduced use of space (d'Orbcastel et al. 2009c; Barrut et al. 2011). Energy costs of airlift pumps for water transport and aeration have been up to 35% lower compared to standard pumps, in particular when used with low head systems (Reinemann et al. 1990; Kassab et al. 2009; d'Orbcastel et al. 2009c).

Oxygenation and aeration

The availability of dissolved oxygen is usually the first factor that limits carrying capacity in RAS; hence the use of oxygenation enables adequate growing conditions, good biofilter performance, and a higher fish biomass in the system. Some systems rely on pure oxygen as the oxygen source while others use aeration to achieve both oxygen addition and carbon dioxide stripping.

The use of pure oxygen can reduce fish production costs by supporting higher fish and feed loading rates at reduced water flow requirements. In turn, it reduces: pump size and cost of pumping; culture tank size or number; size of water reuse equipment; and overall system size. Consequently, the configuration of a RAS determines, to a large extent, the most appropriate type of oxygenation unit for a particular RAS as well as the placement of both the oxygenation and the aeration/stripping units (Summerfelt et al. 2000).

Fish respiration produces carbon dioxide, which is excreted across the gill as CO_2 gas (Colt et al. 2009), while a biofilter also consumes oxygen and generates carbon dioxide. At undesirable concentrations, carbon dioxide may affect fish welfare and reduce water ph. The use of pure oxygen at relatively low specific water exchange rates, requires aeration for CO_2 stripping, the use of chemicals to adjust the pH of production tanks (i.e. adding alkalinity) or a combination of both (Bisogni and Timmons 1994; Grace and Piedrahita 1994; Loyless and Malone 1997).

Pure oxygen gas has been used since the 1970s in order to increase the productivity (i.e. intensifying fish production) and the cost-effectiveness of a RAS (Speece 1981). Nevertheless, providing oxygen to cultured fish may be costly when compared to the cost of feed (Seginer and Mozes 2012), and may be cost-effective only in large scale systems (Sowerbutts and Forster 1980). So the efficiency of oxygenation is important for both technical and economic reasons. In standard temperature and pressure conditions, i.e. 20ºC and 760 mm Hg respectively, oxygenation using pure oxygen as the gas phase could give up to five times the oxygen transfer rate of conventional aeration, in practice 4 times (Petit 1980) and efficiencies recorded for fish culture are considerably higher (Mitchell and Kirby 1976). Operational principles, techniques and equipment for oxygenation has already been well-established (Colt and Watten 1988; Watten 1994) and directions to choose the right oxygenation technology depending on each RAS layout has also been published (Summerfelt et al. 2000).

The choice of selecting one or another will ultimately depend on the economic and technical characteristics of each RAS (Seginer and Mozes 2012). When oxygen is supplied by means of aerators various are the options and efficiencies of design, where surface aerators and packed column aerators are more effective than diffused aeration systems and sub-surface aerators (Hackney and Colt 1982; Loyless and Malone 1998). When aeration is chosen for economy reasons (i.e. reduce equipment cost and usage), the optimal level of dissolved oxygen in the water (i.e. g of O_2/m^3 water) is the lowest permissible (Seginer and Mozes 2012), and this will depend upon reared species and water temperature (Cerezo and Garcia 2004; Cerezo-Valverde et al. 2006).

Rosati et al. 1994 compared 3 types of oxygen and aeration applications in RAS from the technical and economic perspective: (I) liquid oxygen used with a high efficiency dissolution device such as an oxygen column or a U-tube (total energy consumption while generating: 7.69 kWh /kg fish); (II) a surface agitator (28.2 kWh/kg fish) and; (III) a blower with air-stones (65.5 kWh /kg fish). Oxygenation with a paddlewheel aerator (i.e. no pure oxygen supply) is, according to above mentioned authors, the most economical. However, the appropriateness of this method of aeration may be questionable in smaller indoor systems. Thus, for example, aeration energy accounted for around 20% of the total energy consumption in the production cycle (d'Orbcastel et al. 2009b).

Additionally, when capital costs and intensity of production are considered the ranking of these alternate systems may change. The production of liquid oxygen (including the amount of energy needed to produce a unit weight of oxygen and the energy required to transport oxygen to the facility) for a large-scale RAS with temperature control accounts for 0.12% of the total energy used (Colt et al. 2008).

Filtration and/or removal (solids and nitrogen compounds)

Mechanical filtration removes particulate matter, while biological filtration removes dissolved wastes. Typically, a considerable amount of sludge is produced in RAS and this sludge must be treated before it can be disposed of (Losordo and Timmons 1994; van Rijn 1996; Shnel et al. 2002; Suzuki et al. 2003; Timmons and Ebeling 2010). The solids, which are removed as sludge, are composed mainly of fish excretions and uneaten feed, where the volatile (organic) fraction ranges from 50 to 92% (Piedrahita 2003; Gebauer 2004; Gebauer and Eikebrokk 2006; Mirzoyan et al. 2008). Typically, fish sludge is characterized by its low total solid content (1.5–3%) compared to other animal production or industrial wastewater (Mirzoyan et al. 2008). Moreover, waste characteristics may also vary widely, depending on the fish species, feed, management and differences in decay of organic matter within the tanks (Van Rijn 1996).

Solids removal is accomplished by sedimentation, mechanical filtration or centrifugation (Van Rijn 1996). Rotating micro-screens (i.e. drum filters), granular filters and gravity settling units are the most common methods used to remove the solids (Liltvedt and Hansen 1990; Bergheim et al. 1993; Franco-Nava et al. 2004). Nevertheless, up to 95 % of the suspended solids may have a diameter smaller (<20 µm) than mesh size in common filters (30-60µm) and are called ‘fine solids’ (Chen et al 1993) and their removal is accomplished by foam fractionation, chemical oxidation (e.g. ozonation), or biological oxidation. Critical factors in the removal of fine solids are: filtration cycle, particle size, solids loading, and pressure head allowed (Fair et al. 1971; Wheaton 1977; Spotte 1979). The selection of filtration to minimize pressure loss is critical in reducing operating costs, though this may be offset against particle size removal and backwash frequency. Depending on the amounts of solids present, fine solids filters may be used intermittently or on a side-stream to reduce operating costs.

From an energy consumption perspective, mechanical filtration requires energy for backwashing, in addition to providing pumping energy to overcome the head loss through the filter. Normal operating power requirements may be increased up to five times during a backwash cycle, e.g. from 10 to 50 kWh (Csavas and Varadi (1980). Nevertheless, the use of additional air scouring, as an adjunct to water backwash, may reduce the power requirement (Burrows and Combs 1968). As for centrifugal filtration, their efficiency for solid removal is considered poor (Mayo (1976), and besides highly energy intensive (Wheaton 1977); hence its use is not recommended for aquaculture.

Various options are available for nitrification or biofiltration. The choice of a given filter will depend on the strategy taken for the bacterial culture (i.e. suspended growth or fixed film), which as well depends on the strategy used to provide oxygen (Malone and Pfeiffer 2006). Use of suspended growth started in the last two decades (Avnimelech 1999, 2007;

McIntosh 2001; Avnimelech and Kochba 2009), while biological fixed film processes have been used since the early 80s (e.g. Brune and Gunther 1981; Kaiser and Wheaton 1983; Losordo 1991). Hybrid equipment (i.e. systems incorporating aspects of both fixed and suspended-media operation) can also be found.

Within the technologies mentioned, many configurations are used, i.e. moving beds, down-flow filters, rotating biological contactors, trickle, up-flow and fluidized bed filters. Different studies have been published referring to their efficiency in terms of nitrogen removal, specific surface areas and material used (e.g. Chen et al. 1993; Malone et al. 1993; Summerfelt and Cleasby 1996; Kamstra et al. 1998; Eding et al. 2006; Malone and Pfeiffer 2006) but few have mentioned their energy requirements (Sandu et al. 2002).

2.2. Energy source

The peak of the fossil fuel era has already passed and the use of renewable energy sources is expected to increase significantly (Monforti-Ferrario et al. 2015), up to 30–80% in 2100 (Fridleifsson 2001). In Europe, according to the Europe 2020 initiative, renewable energies should account for 20% of the energy produced by the year 2020 (COM 2016). Hydropower and traditional biomass are already important sources in the world's energy mix, contributing about 18% of the total world energy requirements. Meanwhile, the new renewables (i.e. solar, wind and geothermal) contribute only about 2% of the present world primary energy use. In fact, solar energy for electricity production is still not commercially competitive in many places, while biomass, wind and geothermal energy are making relatively fast progress (Fridleifsson 2001).

In this context, in order to find solutions to problems such as the global warming potential, it will be necessary to integrate local energy sources into national/regional systems making use of the most appropriate local and imported energy (Fridleifsson 2001). Therefore, energy conservation at the farm scale (Rosen and Dincer 2001) and replacement of fossil fuels by renewable sources should be supported by industry stakeholders (Aubin et al. 2009).

In the aquaculture literature there are few contributions regarding: (I) the use of renewable energy sources; (II) advantages and/or disadvantages and comparisons between them and; (III) operational costs related to each. Therefore, this section aims at reviewing different energy sources and/or renewable energies used in RAS. Moreover a comparison between different energy sources and their costs is made for different species reared in RAS.

Geothermal energy

Geothermal energy can be used for both electricity and hot water generation for the processing of agricultural products and rearing fish in aquaculture, depending on the temperature and chemistry of the resources. Heat exchangers are often necessary when using geothermal energy due to chemicals in the geothermal waters, such as arsenic and dissolved gases, which are a major problem with regard to plants and animals. The use of geothermal energy in aquaculture is particularly attractive as the temperature range required varies between 25-35ºC (i.e. warm water species) for which there is an abundance of geothermal resources (Lund 2013). The main advantages for RAS are the immediate use of the heat energy to produce electricity, the direct use of geothermal fluid for both heating and cooling (i.e. heat pumps), and the allowance of operating in colder climates (Lund 2013).

The use of geothermal energy in RAS has been extended in countries such as Iceland (Ragnarsson 2014) and Alaska (Ogle ,no year available). In Egypt, catfish production was achieved by using geothermal energy handled by a plate heat exchanger in a RAS (Farghally et al. 2014). Fish breeding using geothermal energy has been also successful in Japan, China, and the United States. Tilapia, salmon and trout are the most common species, but tropical fish, lobsters, shrimp, and prawns are also being farmed with geothermal energy farms (Ragnarsson 2014).

Solar energy

Solar technologies are twofold depending on how they capture and distribute solar energy or convert it into solar power (Fuller 2007): passive solar (i.e. natural convection and direct solar absorption by the water body) or active solar (i.e. solar collector such as photovoltaic systems). In aquaculture, both active and passive technologies have been used so far. Contributions include: using passive technologies (Brown et al. 1979; Van Toever and Mackay 1980; Yuschak and Richards 1987; Provenzano and Winfield 1987; Shilo and Sarig 1989); and using active technologies (Ayles et al. 1980; Ray 1984; Plaia and Willis 1985; Fuller et al. 1998).

The use of solar energy decreases the reliance on fossil fuel of a RAS, with those farms requiring a considerable heating contribution showing the highest benefit-cost ratio. There are a few examples using solar panels: (I) an experiment in the Canadian hatchery industry for Atlantic salmon and rainbow trout production evaluated the economic viability of various water heating techniques (including oil, gas, electricity, propane, solar, and combination systems) (Carpenter 1993); (II) a simulation model using a greenhouse with and without solar collectors (Fuller 2007) and; (III) an experimental RAS project designed to rear 432 kg of trout in Canada which depended on the use of solar collectors for water temperature maintenance. Here, coupling a solar collecting system (i.e. 91 % of the required heat by solar panels) to fish rearing units decreased the total energy consumption from 14.25 to 2.31 E-03 kWh/kg fish. At a commercial scale, in Canada, solar heating was integrated with a conventional propane heating system, saving around 11,500 € per year (Toner 2002).

Waste heat from industry

Waste heat from industry has been used for commercial oyster, penaeid shrimp and salmon farming; thermal effluents in culturing American lobster and; the use of thermal waste water to produce catfish in Pennsylvania (Rickard, 1998). Eel and salmonids fingerlings productions using heat from power plants (Lemercier and Serene 1980;

Ingebrigtsen and Torrissen 1980, respectively) and salmonid culture using hydroelectric waste heat (Sutterlin 1981; Mercer 1984) are other examples. Positive results of using waste heat from thermal electric or hydro-electric power station were obtained and it was concluded that it may offer substantial energy and cost savings to salmon aquaculture in Canada (Mercer 1984). Additionally, animal growth was satisfactory when using waste heat and water from zero discharge power plants in the Great Basin (Heckmann et al. 1984). Nevertheless, the use of waste heat has not been widely extended within the industry as there may be significant problems such us hygiene issues due to its usage. Herein, there are not updated examples or references in the bibliography regarding this type of energy source.

Other renewable energies

Hydropower is a renewable energy source based on the natural water cycle and it is the most mature, reliable and cost-effective renewable power generation technology available (Brown et al. 2011); indeed the only large scale and cost efficient storage technology available today (IRENA 2012). Hydropower (i.e. as part of the energy source from the mix in the grid available where the electricity takes place) has been successfully used in RAS, decreasing environmental impacts and economic costs (Liu et al. 2016).

Few studies have been published about the use of biomass, wind power or tidal energy in RAS. The latest report about the potential for renewable energy usage in aquaculture presented a case study about a marine finfish RAS facility producing 200 t of turbot/halibut (Toner 2002). It was concluded that wind and wave power may be viable sources given the energy demand (13,767 kWh/ week). The installation of those systems would require a large capital outlay but this could be recouped within a period of about six years.

2.3. Energy consumption and energy sources in different RAS

The number of RAS farms around the world is steadily increasing (Martins et al. 2010; Badiola et al. 2012, 2014; Dalsgaard et al. 2013). This is reflected in the latest publications and in some worldwide research made by the authors through personal communication and social networking. Data compiled in the research is reflected in Figure 3.1 (i.e. worldwide countries ranged according to the number of RAS companies) and Table 3.2 (i.e. Europe's fish production in RAS between 1986 and 2014). In the US and Europe, the number of RAS installations is around 360 (USDA 2013 Census of Agriculture; Badiola et al 2014). Norway and Canada represent important RAS industry countries, mainly for salmon production (Dalsgaard et al 2013), while China is increasing its yearly production with the construction of new, large indoor RAS facilities (Murray et al 2014). Salmon, tilapia, trout, eel, turbot, catfish and shrimp represent the main species farmed (Badiola et al 2012). This increased number of RAS farms around the word inherently implies the use of energy and its consequences both for companies (i.e. economic) and the environment (i.e. regional and global).

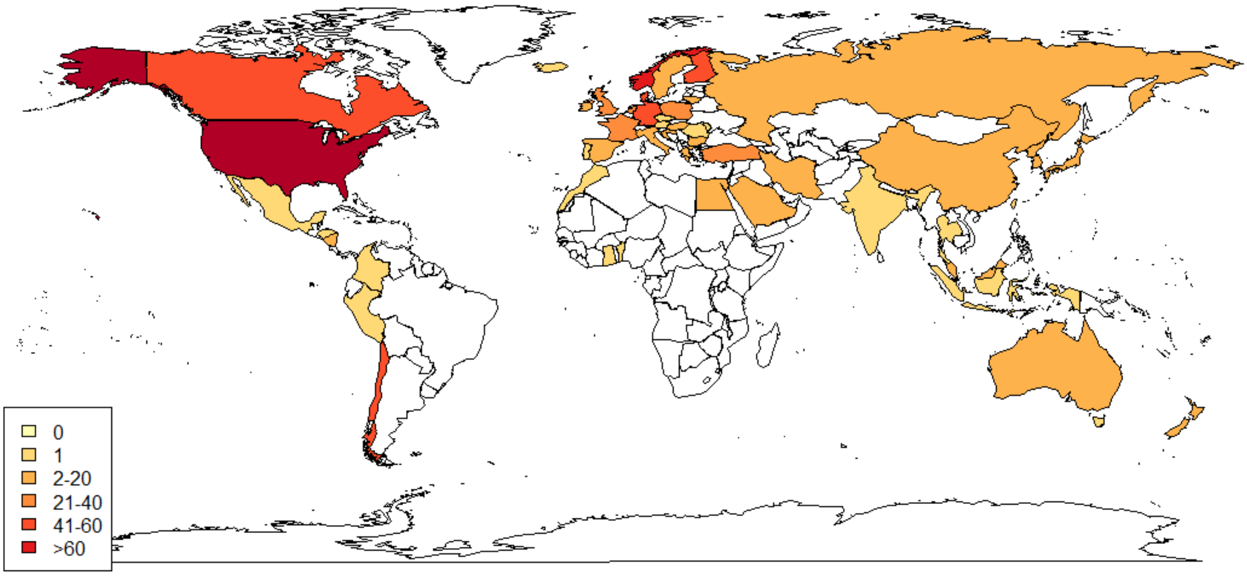


Fig. 3.1 Worldwide countries ranged according to the number of RAS companies in each country. Information updated from Martins et al. 2010; Badiola et al. 2012, 2014 and Dalsgaard et al. 2013 after a worldwide research made by the authors through personal communication and social networking during the last 4 years.

A possible parameter used when comparing RAS systems, is the energy consumption index (i.e. kWh/kg fish). It differs by species and RAS as it depends on factors such as location and production volume (Table 3.2). Overall, the range varies widely between 2.9 and 81.48 kWh/kg. Reasons for such difference may be due to the rearing stage (e.g. Colt et al. 2008 and Liu et al. 2016, smolts and grow-out, respectively; objectives of the study), other design parameters (fully recirculated and or partial reuse systems (Summerfelt et al. 2009)) or technical choices for the regulation of the temperature (e.g. Aubin et al. 2009). A survey compared RAS and flow-through systems (FTS) from Norway, Canada and Iceland (Bergheim et al. no year available). The energy used for production was similar in the Norwegian RAS and the average value for Icelandic FTS farms. However, in two of the Icelandic farms, water flowed by gravity through the tanks, i.e. no energy was spent on pumping water. Therefore, when those farms were excluded from the mean, the average energy used in the Icelandic FTS was 7.6 kWh/kg fish, similar to values reported for Norwegian RAS and the Norwegian and Icelandic FTS systems (Summerfelt et al. 2004). In the Canadian RAS, the water was only aerated while recirculated but not oxygenated, which would explain the increased amount of energy used compared with the Icelandic FTS and Norwegian RAS. Similarly to the Atlantic salmon smolt production in Table 3.2, where high amounts of oxygen were required (i.e. hatchery and smolt production stages) and supplied by liquid oxygen.

The comparison between systems should not be generalized and assumptions taken should be specified. As previously mentioned, each system is different and dependent on several factors. Thus, most of the times it is very difficult to know the factors included in the studies, resulting in very different values.

Carbon footprint

Most of the studies show that the preferred energy sources are fossil fuel based (i.e. coal and natural gas) with increased CO_2 emissions in comparison to renewable sources. In contrast, renewable energies clearly may decrease the greenhouse gas emission: e.g. 4.86 kWh/kg from hydropower emitted 3.73 kg CO_2-eq, while 0.54 kWh/kg from coal energy emitted 7.01 kg CO_2-eq (Liu et al. 2016). Nevertheless, as in the case of trout farming in France (i.e. nuclear based energy), CO_2 emissions were much lower than using fossil fuel based electricity resulting in higher environmental impacts (i.e. eutrophication potential and water ecotoxicity) (Aubin et al. 2009). Therefore, the location of the farm is an important parameter and may change created impacts. Nevertheless, in general, RAS companies' electricity is generated in a public utility, limiting the options of the energy source. In this manner, the unique choice would come if the company decides to generate the electricity independent from an utility company (i.e. when a public utility is unavailable or unreliable).

Figure 3.2 shows the results from a comparative study and a sensitivity analysis made by the authors of three different contributions taken from the literature using fossil fuels. Firstly, CO_2 emissions were calculated for each study, taking into account the country of location. This was the first scenario (i.e. F). After, for the sensitivity analysis, different hypothetical scenarios were created varying (in terms of %) the source of energy used in each study: (I) 20/80- fossil fuel/geothermal energy (FG); (II) 50/50 –fossil fuel/wind power (FW) and; (III) 10/90- fossil fuel/hydropower (FH). Note that these are hypothetical scenarios including assumptions taken by the authors. Thus, in the reality, the reasons for the changes in the outcome may vary. Finally, energy-related operational costs were calculated for all four scenarios. Data sources were gathered from the bibliography and current statistical websites detailed in the figure.

Table 3. 2. Literature values for species, country, production volume, harvest weight, energy source and energy consumption of various cultured products per live-weight kilo at farm gate.

Species	Country	Production volume (t)	Harvest weight (kg)	Energy source	Energy consumption (kWh/kg fish)	Reference
Turbot	Brittany (France)	70	1.2	Fossil fuels	81.48 [a]	Aubin et al. 2009
Artic char	Nova Scotia	46.2	1.5	Fossil fuels (77% coal)	22.6	Ayer and Tyedmers 2009
Turbot	Galicia (Spain)	3,500	1	Fossil fuels	20.03 [b]	Iribarren et al. 2012
Atlantic salmon smolts	Pacific Northwest (US)	192	-	Fossil fuels (98 % natural gas)	80.64	Colt et al. 2008
Trout (FCR 0.8)	France	478	-	86.6% nuclear energy	16.14	d'Orbcastel et al 2009b
Trout (FCR 1.1)	France	478	-	86.6% nuclear energy	17.70	d'Orbcastel et al 2009b
Rainbow trout	Denmark	1	-	Fossil fuels	19.6	Samuel-Fitwi et al 2013
Rainbow trout	Iran	1,000	-	Fossil fuels (80% natural gas)	8.1	Dekamin et al 2015
Atlantic salmon	US	3,300	-	90% hydropower 10% coal power	5.4	Liu et al 2016
Florida Pompano [c]	Florida (US)	0.43	0.6	-	40.3	Pfeiffer and Riche 2011
Atlantic cod	Basque region (Spain)	-	1	Fossil fuels	29.43	Badiola et al 2016
Sea bass	Tunisia	2,500	0.4	Fossil fuels	49.16[d]	Jerbi et al 2012
Sea bass	Tunisia	2,500	0.4	Fossil fuels	78.40[e]	Jerbi et al 2012
Atlantic salmon smolts[f]	US	11.246	137	-	19-26	Summerfelt et al 2009
Rainbow trout[g]	US	2.505	103	-	2.9	Summerfelt et al 2004

[a] takes into account feed production (11%), equipment (1%), infrastructures (1%), chemicals (1%) and energy consumption in the farm (86%)

[b] hatching and nursing (14.84), growing (3.15), on-growing and final operations (2.04)

[c] no information is provided about the energy source used

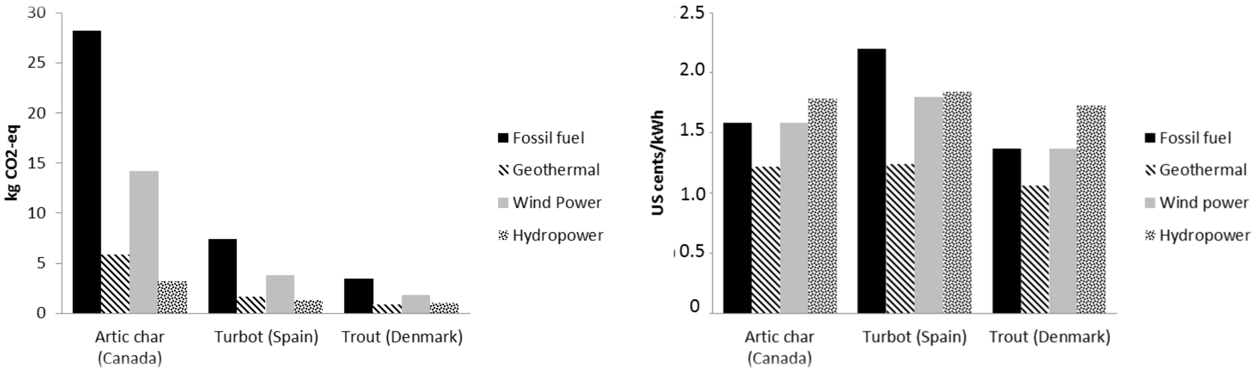
[d] production system based in a hatchery working as a RAS and traditional raceway for the grow-out stage

[e] production system based in a hatchery working as a RAS and cascade raceway for the grow-out stage.

[f] partial-reuse system (i.e.87-89% water treated)

[g] partial-reuse system (i.e. 80-85% water treated)

From the emissions perspectives, there is a general linear decrease with the % of renewables incorporated in three of the cases. From an economic point of view, results are more variable but the implementation of renewable energies seems feasible in three of the countries. It is important to remark that such comparisons (Fig. 3.2) are simulations based on assumptions and average values from the literature (i.e. cost of energy and kg CO_2-eq/kWh). Therefore, the reality may differ by country as energy sources are very site-specific, creating diverse environmental impacts (both in quantity and severity). In geothermal development for the generation of electricity for example, about 50% of total costs are related to the identification and characterization of reservoirs which greatly varies between countries, affecting the total costs (Barbier 2002). Moreover, wind power installations for example may be onshore or offshore directly impacting CO_2 emissions (3.00E-03 to 4.50E-03 or 7.00E-03 to 2.30E-02 CO_2-eq, respectively) (Thomson and Harrison 2015).



		US cents/kWh	Reference	kg CO_2 eq /kWh	Reference
Fossil fuel	Canada	0.07	The Statistic Portal 2015	1.248	Ayer and Tyedmers 2009
	Spain	0.11	The Statistic Portal 2015	0.372	IDEA 2014
	Denmark	0.07	The Statistic Portal 2015	0.176	Ecoinvent
Geothermal	Canada	0.05	Fridleisson 2001	0.01	Barbier 1997
	Spain	0.05	Fridleisson 2001	0.01	Barbier 1997
	Denmark	0.05	Fridleisson 2001	0.01	Barbier 1997
Wind Power	Canada	0.07	Fridleisson 2001	0.0092	Warner and Heath 2012; Whitaker et al 2012; Thomson and Harrison 2015
	Spain	0.07	Fridleisson 2001	0.0092	Warner and Heath 2012; Whitaker et al 2012; Thomson and Harrison 2015
	Denmark	0.07	Fridleisson 2001	0.0092	Warner and Heath 2012; Whitaker et al 2012; Thomson and Harrison 2015
Hydropower	Canada	0.08	IRENA. 2012	0.02	Raadal et al 2011
	Spain	0.09	IRENA. 2012	0.03	Raadal et al 2011
	Denmark	0.09	IRENA. 2012	0.04	Raadal et al 2011

Fig. 3.2 Comparative study of 3 different studies analyzing: CO_2 emissions by different energy sources and operational costs created by the energy consumption in each of them.

3. Stakeholders' vision:

The opinions of stakeholders are critical for the advancement of any industry and/or company. They provide the closest judgment from the consumers which are in last term the ones dictating either success or failure. A survey (Annex 3.1) was conducted to analyze the perspective of the industry regarding energy use in RAS. The main objective was to investigate how the energy within the system (i.e. knowing which are the most energy consuming devices) is used; which is/are the energy sources (i.e. if a renewable source is used); and how much energy is used to produce the final product (i.e. kWh/kg). This would help to identify priorities for future research in order to reduce both the environmental and economic impacts of RAS. Furthermore, the analysis considered the priorities of the industry in terms of investing or not to enhance their sustainability and which energy (i.e. costs) saving measures were applied. More subjective viewpoints and experiences of the researchers and consultants would help compare and contrast diverse ideas and approaches for the future. Survey respondents were asked about which parameters influence the energy use in RAS and which types of designs would help to enhance the efficiency of the overall system. The questions were taken as a baseline but and could be modified depending on the interviewee´s expertise.

The questionnaire was developed for both fish farms (i.e. RAS producing farms) and producers and opinions about personal experiences were collected from researchers, consultants and manufacturers. In the framework that new technologies are gaining more importance, a wide range of communication channels (i.e. social networks, personal communication, and interviews) were used to reach different interviewees.

In total, 96 people were contacted directly or through social media like LinkedIn and Facebook. After seven months of contacting people, only 10 questionnaires were returned from the industry and comments from 15 people from both university and research centers were received. Respondents were from Australia, Belgium, Bulgaria, Finland, France, Germany, Israel, Norway, Slovenia, South Africa and USA. Species reared by survey responders included: Atlantic salmon (*Salmo salar)*, pangasius (*Pangasius buchanani)*, clarias (*Clarias anguillaris)*, Arctic char (*Salvelinus alpinus)*, pikeperch (*Sander lucioperca)*, tilapia (*Oreochromis niloticus)*, rainbow trout (*Oncorhynchus mykiss)*, and sturgeon (*Acipenser naccarii*). The choice of such species was because they are high value and robust for RAS; market's convenience; fast growers (i.e. tilapia) and; internationally proven species.

RAS farms differ from each other in the design, as this is dependent on the location, available resources, and biological requirements of the species reared. However, basic procedures such as monitoring certain parameters are common to all systems. Thus, in all ten farms, dissolved oxygen, temperature and pH were continuously monitored, while CO_2, TAN, NO_2 and NO_3 were measured about once per week. In cases where the energy was measured, it was done as a total value for the whole system and not for each piece of energy consuming equipment. In all cases, production buildings were isolated and two of the respondents reported covering the tanks for heat saving purposes. Most systems (80 %) used oxygenation instead of aeration and CO_2 was removed by some form of aeration. Electricity was obtained from the local grid in 60 % of the cases and renewable energies such as solar energy (in South Africa), biogas from a local wetland (in Finland), wind power (in Sweden) and energy from a hydroelectric plant (in Norway) were mentioned.

In relation to energy recovery systems designed/applied, one of the respondents reported exchanging heat between the incoming (i.e. make-up water) and outgoing water through a heat-exchanger. Other answers were: retaining heat based on the system's operation/water use; controlling the energy use of CO_2 stripping through pH/CO_2 set-points for on/off control of blowers for energy saving and; increasing the recirculation rate through the use of denitrification technologies which resulted in energy use reduction and cost savings. Moreover, using the system's sludge for local farming purposes and producing energy for other nearby companies through a bioreactor supplied by sludge, guts from the processing stage and mortalities, were also mentioned.

Among the respondents, RAS were considered an "environmentally friendly" fish production method mainly due to: less water usage from the environment compared to other culture technologies such as flow through systems; decrease of the eutrophication potential of the outgoing water; elimination of potential disease transfer and genetic contamination of wild stocks; use of no or very little vaccines or antibiotics because of a biosecure culture environment and the possibility of reusing discharged nutrients in agriculture. Nevertheless, in practice, sustainability of the systems (i.e. economic and environmental) was considered to be uncertain and the use of energy and its environmental impact was of no concern to the respondents. In fact, concerns identified by responders included (Fig. 3.3): identifying alternatives to fishmeal (35%); enhancing animal welfare (i.e. increased biomass production, increased survivals and reduced maturation with the subsequent decrease of product downgrades) (26%); decreasing the feed conversion ratio (23%); decreasing the use of chemicals (11%) and decreasing the use of energy and thus, created environmental impacts (5%).

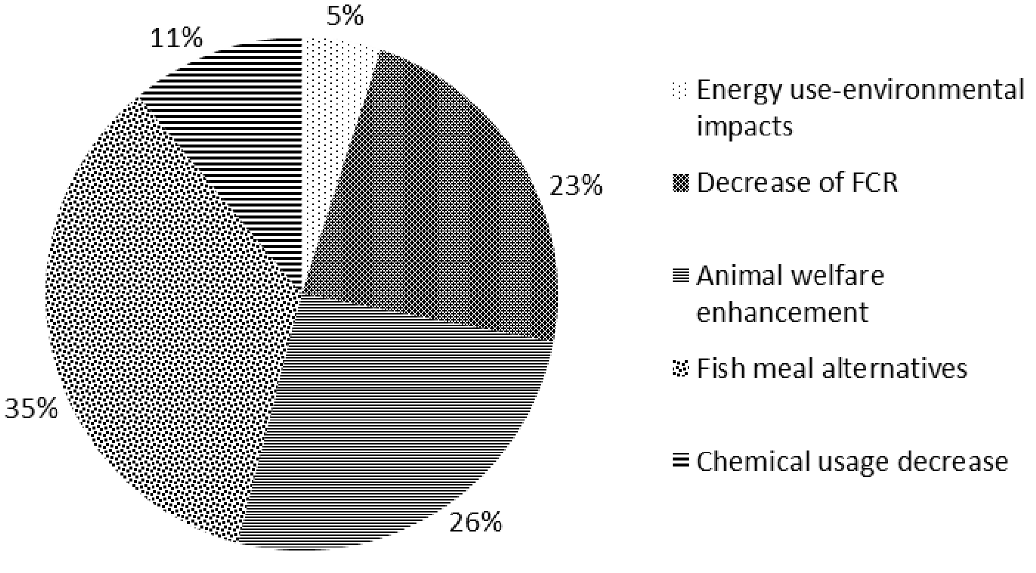


Fig. 3.3 Concerns around RAS production identified by responders of the present study.

4. Designing a RAS: towards an efficient system

The following section aims to present an optimized RAS design approach including: optimized RAS unit processes, engineered system integration and engineered equipment selections. Moreover, alternative design solutions for each system and subsystem and component are provided. Optimized solutions or alternatives given below are from authors own experience and opinion made after the study.

Setting up a RAS requires that considerations of costs, fish welfare and product quality be taken into account. Increasingly, it also involves minimizing the potential environmental impacts. Creating and/or designing an energy efficient production RAS will help save money and energy, which will inherently help achieve a sustainable (i.e. environmental, social and economic) production operation.

The design of a RAS should ensure a proper balance of the important parameters affecting water quality and fish productivity. Important general water quality parameters for cool and warm water species include water temperature, oxygen, carbon dioxide, total suspended solids, total ammonia, unionized ammonia, nitrite and nitrate. Thus, a mass balance should be done on all of those variables (i.e. at steady state: transport in of “x” + production of “x” = transport out of “x” + consumption of "x", where “x” is the studied variable) (Timmons and Ebeling 2010). Fig. 3.4 shows the relation between a general mass balance on a fish culture tank and the treatment device afterwards. The concentration of any of the parameters leaving the treatment device can be easily solved since the water flow in and water flow out are equal. In such manner,

$$C_{out} = C_{in} + T/100\ (C_{best} - C_{in})\quad \text{(eq 1)}$$

where C_{out} is the outgoing concentration of a given parameter (e.g. mg/l); C_{in} is the incoming concentration (e.g. mg/l); T is time (days); and C_{best} is the absolute best result obtainable by the treatment device (Timmons and Ebeling 2010).

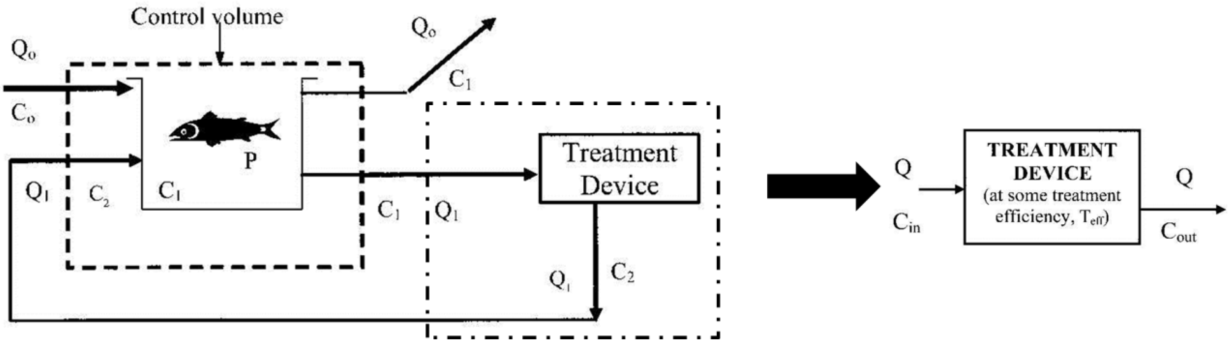


Fig. 3.4. A general mass balance of a production tank and a general treatment device (Timmons and Ebeling 2010).

As discussed in Section 2 (Energy use in RAS), various unit processes (i.e. solids and waste solids removal, aeration or oxygenation, removal of nitrogenous compounds, carbon dioxide removal) and components (e.g. filters, biofilters, air stones, pumps) are used in RAS. However every RAS is different and factors such as location, species and production volumes would directly affect the overall design (Badiola et al. 2012).

Table 3.3 presents a relation between water quality parameters, unit processes and design issues. Candidate technologies, systems and equipment are related to each other.

Table 3.3. Relation between water quality parameters, unit processes and design issues. Candidate technologies, systems and equipment relation with each other.

Important Water Quality and system's general parameters							
Settleable and total Suspended Solids	Ammonia and nitrite	Carbon Dioxide (CO_2)	Primary and secondary recirculated flow	Dissolved oxygen	Dissolved and fine solids	Bacterial species and colony counts	Temperature
Unit Processes							
Waste Solids Removal	Biofiltration (Nitrification)	CO_2 removal	Pumping	Aeration and/or oxygenation	Dissolved and fine solids removal	Disinfection	Heating and cooling
Key Application Design Issues							
Minimize at the source	Critical to fish health and growth performance. Must be robust and user friendly	Empirical design, limited commercial equipment	Must couple pump selection with RAS hydraulic profile	Critical process	Important to fish health and growth performance	Important to fish health and growth performance	Optimized for fish growth performance
Candidate Technology, Systems and Equipment							
Settling basin	Packed column	CO_2 stripper with and without	Swimming pool	Surface aerators	Foam fractionator	Ultraviolet	Electric resistance

Radial flow separator	Trickling filter	packed media	pumps	Air stones	Suface skimmer	disinfection	heat
Particle trap	Rotating contactor	Surface aerator	Centrifugal pumps	Fine pore diffusers	Bead filter	Ozonation	Plate type heat exchangers
Microscreen drum filter	Bead filter	Moving bed reactor	Axial flow pumps	Contact columns & cones	Nutrient limited biofiltration	Ozone fed foam fractionators	Shell & tube heat exchangers
Bead filter	Fluidized sand		Air lift pumps	U-tubes			Geothermal heat pumps & chillers
Double drain tank	Moving-bed			Low head oxygenator			Water-cooled heat pumps & chillers
Mixed rearing cells	Micro-bead			Hooded agitators			Air-cooled heat pumps & chillers
				Liquid oxygen (LOX)			Energy recovery
				PSA & VSA generators			

Settleable and total suspended solids

The design goal should be to minimize the presence of solids within the system. Solids are the source of most water quality problems and impact the efficiency of most other treatment devices (Badiola et al. 2012). In fact, the presence of solids can stress the bacterial community (Malone and Pfeiffer 2006; Emparanza 2009), hampering for example biofilter (Singh et al. 1999) and ozonation (Summerfelt et al. 2009a) performance. The increase of ammonia concentrations due to a less efficient treatment will increase the energy demand of the system as its capacity will be affected. Thus, based in the research undertaken, a rapid and efficient removal of settleable and suspended solids could be achieved using a double tank drain with a radial flow separator and a microscreen filter.

Ammonia and nitrite

Ammonia and nitrite are critical to fish health and growth performance. Biofilter characteristics determine the maintenance requirements as well as the management techniques required in the production (Badiola et al. 2012). In this manner, for example, a parameter imbalance due to daily procedures (e.g. a rapid fluctuation of the ammonia or nitrite concentration during feeding hours) and/or biomass variation can affect biofilter's efficiency. A variety of biofilters is available commercially. For larger systems, low-head, efficiently aerated moving-bed bioreactors are the prevailing choice, while micro-bead bioreactors are a competitive, lower cost alternative (Timmons et al. 2006; Fadhil et al. 2011).

Carbon dioxide

The prediction of carbon dioxide removal rate may be difficult due to diverse factors involved (Hu et al. 2011). Currently, there is limited availability of commercial equipment for CO_2 removal. Conversely, CO_2 strippers with or without packed media, surface aerators or moving bed reactors are candidate technologies. When coupled with moving-bed or micro-bead biofilters, surface aerators provide the additional aeration and the CO_2 stripping required (Liu et al. 2013). Optimized solutions include surface aerator with variable frequency control.

Water pumping

Pump selection must be done to match the RAS hydraulic profile. There are various types of pumps available (i.e. centrifugal, axial flow, air lift). In general, axial flow pumps can be more hydraulically and energy efficient. Properly selected and trimmed, low-head centrifugal pumps are needed for higher head systems. Furthermore, variable frequency control is an alternative to trimming impeller. Nevertheless, in a real production, pump selection is highly dependent on flow rate and/or head requirements. Additionally, the availability of pumps to match required flows may be limited. Thus, it is difficult to recommend a single type of pump. An optimized solution, resulted from this study, may be an axial flow pump with variable frequency control.

Dissolved oxygen

There are many candidate technologies available for oxygen addition and oxygen and electrical power costs are site specific. The only way to determine if oxygenation is cost effective is to do a detailed cost/benefit analysis. The question is whether the cost of installation and running of an oxygenation system is offset by the extra fish that can be grown during the service life of the entire system. In other words, the cost per kg of fish of oxygenation is compared to the reduced cost per kg of system depreciation (e.g. on a moderately large system, oxygenation can add about 5% to the cost per kg and can be determinant to be justified). Optimized solutions are: (I) U-tubes; (II) contact cones on side-stream pumps with variable frequency control and; (III) site specific liquid oxygen or generator selection.

Dissolved and fine solids removal

Dissolved and fine solids are important to fish health and growth performance although the implementation of a specific device for their removal is not always needed. An optimized solution for an effective removal would be a robust biofiltration together with ozone fed foam fractionator.

Bacterial species and colony counts

Water quality has to be optimum for fish health and growth performance which includes achieving disinfected rearing water. The high stocking densities, associated fish stress and increased nutrient loads found in RAS create an ideal environment for fish pathogens. Diverse are the steps taken to reduce the risk of disease outbreaks in RAS: (I) the use of standard quarantine procedures for any fish introduced (prior entering production tanks); (II) reduce the pathogen load introduced via the source water treating the make up water and; (III) the disinfection of effluent waters before introduction to the environment to prevent the translocation of exotic diseases.

Some type of disinfection is usually employed such as ultraviolet disinfection units and/or ozonators where a significant level of disinfection is achieved (Kingsley et al. 2008). The use of disinfection procedures is based in rearing species or life stages under production. An optimized model or solution would be the use of ozone fed foam fractionators where bacterial reduction achieved is moderate and bacteria are physically removed by the fractionator (Phillips et al. 2004). Nevertheless, the use of disinfection as part of the recycle loop should be applied in specific situations as it could be counterproductive as general use.

Temperature

Heating and/or cooling of the rearing water is achieved by different equipment: electric resistance heaters, plate type heat exchangers, shell and tube heat exchangers, geothermal heat pumps and chillers, water-cooled heat pumps and chillers, air-cooled heat pumps and chillers, energy recovery. Apart from this, site selection has tremendous cost implications in temperature's control, energy costs and shipping costs directly linked with the species produced.

5. Concluding remarks:

RAS designs are being developed and improved, incorporating new technologies (e.g. Piedrahita et al. 1996; van Rijn 1996; Cripps and Bergheim 2000; Summerfelt and Penne 2005; Eding et al. 2006; Summerfelt 2006). However, studies describing new technologies typically do not include considerations of energy use by the technologies or of their impact on total energy consumption and system efficiency (Badiola et al. 2012). In the recent past, the statement "sustainable production", did not necessarily include energy use considerations (e.g. Crab et al. 2007; Tal et al. 2009). Nevertheless, excessive energy use generates significant economic and environmental impacts. Thus, in order to emphasize the advantages provided by RAS, energy consumption should be minimized relative to production (i.e. kWh/kg fish produced).

The challenge for designers is to develop systems that minimize production cost per unit cost of production (including capital and operational costs). Optimal system configuration, from economic (i.e. pumping cost minimization) and environmental points of view, have yet to be defined and studied according to each farming context (i.e. the energy use due to feed, electricity and oxygen consumption is system-dependent). According to the specific context of the farm, a compromise has to be found between water dependence, waste emission, energy consumption and productivity in order to orient the system towards economic and environmentally sustainable production.

In such context, and in accordance to the extensive literature review and interviews made by the authors of the present contribution, energy use in RAS could be reduced by:

- Investing in an area where on average the optimum environmental conditions (e.g. temperature) are naturally available.

- Meeting overall needs of the species of concern while minimizing energy costs.

- Improving both the system design and management of airlifts and bio-filters. Finding a compromise between: an optimal design for water circulation and water oxygenation of the airlift and the backwash and operation of the bio-filters.

- Minimizing height differences between RAS compartments, i.e. low head RAS (RAS should be designed to avoid lifting of the water, when possible).

- Land or building prices may outweigh the advantage of spreading out horizontally. However, more "vertical" systems may come at the expense of higher pumping costs.

The electricity generation obtained from fossil fuels causes local and global environmental problems (e.g. CO_2 emissions). Thus, the use of renewable energy sources on RAS farming, companies needs to be thoroughly assessed for its suitability in each particular situation. An economic analysis needs to compare the cost of connection as well as the use of alternative sources, considering: the consequences of power outages; the fact that a facility is not totally relying on an intermittent renewable energy source and; the accessibility of a possible back-up. Thus, production’s audits including Life Cycle Assessments and integrating energy audits would be the way towards a cost-effective industry (Badiola et al. 2017).

Appendix 3.1 RAS survey

This is part of a PhD research. The core goal of the project is to analyze the economic and environmental sustainability factors of Recirculating Aquaculture Systems (RAS), e.g. energy use (kWh/kg); different energy sources. We hope that learning about: (I) how energy is used within the system (i.e. knowing which of the devices the most energy consuming are); (II) which the energy sources (i.e. if any renewable source is applied) is/are; and (III) how much energy to produce the final product (i.e. kWh/kg) is used, will help to identify priorities for future research in order to reduce both the environmental and economic impacts of RAS. Moreover, to analyze the priorities of the industry in terms of investing or not to enhance their sustainability and which are the energy (i.e. costs) saving measures applied. This information will aid to understand where improvements can be made that would benefit the industry. The assessment is limited to the farm and so, feed production, juveniles (if applied), oxygen production and their transportation are not taken into consideration (although it is known that they are also economically costly). All information provided will be treated as confidential.

1- Which is the production (kg/year)? Which species? Why was it chosen?

2- Which is the difference between the culture temperature and the temperature outside the tanks (i.e. inside the building)?

3- Is your building insulated? Which is the temperature difference between inside and outside the building?

4- Which is the water exchange ratio?

5- Do you have an expansion tank were you hold certain amount of water like a back-up?

6- Which parameters are continuously monitored in the system? How?

7- How is oxygen and CO_2 transferred within the production?

8- Do you know how much kWh/kg is used? Do you measure the energy used in the entire system or do you motorize each of the devices?

9- Which energy consuming devices are within the system? Do you consider is it a high-tech system?

10- Do you think that high-tech is synonym of sustainable?

11- Which energy sources do you use? (i.e. gas, electricity, renewable energies included...). If you are using renewable energies, why did you choose that one?

12- Which is the cost of the kWh, $/kWh? List sources and unitary costs.

13- Do you measure the consumption fluctuations? And do you adequate the source and the consumption of energy according to the national energy plan rate (if there is any)?

14- Do you have any energy recovery system (e.g. use of wastes for generating energy)? Do you have an energy plan which changes between the peak consuming hours and the rest of the hours? Are you involved in any energy saving measure plan?

15- Is the energy for the operational requirements the most costly item? If not, which one is it (%)? (e.g. feed, labor, energy, maintenance, expenses as probiotics, water, wastewater)?

16- Have you made any variation in the system or RAS operation in order to decrease the energy consumption?

17- Will you invest more in technology if this would decrease the environmental footprint of your system gaining image of the company? Marketing improvements.

18- Which performance indicators are important from your point of view or the ones you take into account in order to explain the viability (economically speaking) and the sustainability of your company?

19- What does the statement “RAS are environmentally friendly” mean to you? What does include (water pollution, escapees, less water consumption…)?

Contribution 4

Integration of energy audits in Life Cycle Assessment methodology to improve the environmental performance assessment of Recirculating Aquaculture Systems.

This Contribution was submitted on the 16th March 2016 to:

Badiola, M., Basurko, O.C., Gabiña, G., Mendiola, D. (2017). Integration of energy audits in Life Cycle Assessment methodology to improve the environmental performance assessment of Recirculating Aquaculture System. *Journal of Cleaner Production* (under review)

Summary

In Recirculating Aquaculture Systems (RAS), water is continuously treated and recirculated as opposed to being discharged untreated into the environment as in other type of fish production systems; the design and production parameters will determine the overall energy consumption. This energy-intensive nature hampers their sustainability and cost-effectiveness. This paper proposes a combination of two methods (i.e. Life Cycle Assessment (LCA) with energy audits) to: improve environmental performance of RAS, identify energy consumption and thus, its environmental and monetary effects in order to seek cost reduction. The proposed methodology was proved with a case study focused in a pilot-scale RAS unit used in codfish *(Gadus morhua)* production, located in the Basque coastal area (northern Spain). Feed and juvenile production/transportation, oxygen transportation and energy consumed during the whole experiment were considered as inputs for the assessment. Energy consumption was measured both continuously by an energy meter embedded in the RAS unit as well as with a portable energy analyzer to measure each of the energy-consuming devices independently. Although the system required an average of 29.40 kWh/kg fish for successful system operation, the energy consumption varied by season presenting maximum and minimum periods of 40.57 and 18.43 kWh/kg fish, respectively. Main consumers included the heat pump, followed by the main and secondary pumps, respectively. Energy audit’s results show the success in identifying the devices that consumed the largest amount of energy, and recorded data served to feed the Life Cycle Inventory and perform a more complete and precise LCA. Fossil fuel based on-farm electricity for the on-growing of fish was shown to be the most environmentally unfriendly input; it was the major impact producer in the assessed impact categories. It showed a temporal variability depending on the water temperature, which resulted to be the main factor linked to the energy use. This aided performing a precise assessment including system-specific scenarios. The combination of LCA and on-farm energy audit represents a useful tool to secure a more complete assessment with a periodic assessment to design a less energy intensive, profitable and sustainable system; likewise, it increases the speed and transparency of governance and decision-making, taking into account the time-based fluctuation of the energy consumption throughout the production cycle.

1. Introduction

Water pollution is one of the biggest challenges European aquaculture is facing (AQUAeTREAT 2003). Thus, current policies created for aquaculture's development highlight the need of an industry that minimizes its impact on the environment (COM 2002; COM 2009); in this scope Recirculating Aquaculture Systems (RAS) are proven to be a viable solution (Masser et al. 1999; Timmons and Ebeling 2010; Martins et al. 2010; Dalsgaard et al. 2013). RAS started to develop in the 70s based on sewage treatment plants (Asche 2008). RAS are technologically advanced systems, where several devices treat the water in order to achieve the right parameters for fish to be reared. They are designed specifically to: reduce the amount of water required and waste produced from traditional flow-through systems (known as raceways or tanks where the same amount of water is taken and discharged) (Blancheton 2000), isolate the culture environment from surrounding ecosystems reducing the proximate ecological impacts (i.e. surrounding water bodies pollution, habitat interactions) typically associated with more open production systems, such as net-pens and raceways (Ayer and Tyedmers 2009), and ensure the prevention of inclusion of pathogens guarantying chemical-free productions (Badiola et al. 2014). Nevertheless, RAS are up to 1.4-1.8 times more energy intensive than traditional flow-through systems (d'Orbcastel et al. 2009b), fact that hinder their environmental sustainability. Moreover, in the last year, more efficient products to reduce energy and resource consumption are on demand, requiring the improvement of the energy efficiency and eco-design of products (COM 2016). Hence, on-farm energy use (i.e. fossil energy) should be also quantified (i.e. time-based quantification) and taken into account when eco-designing and/or assessing their design and operations for further development of the RAS industry and increased production volumes from these systems (Ang et al. 2010).

Life Cycle Assessment (LCA) is generally accepted internationally as a strong tool for providing inputs to be considered while assessing the environmental sustainability of a product or process, including those of aquaculture such as salmonid feeds (Papatryphon et al. 2004; Boissy et al. 2011), characterization of turbot farming (Iribarren et al. 2012), the carbon footprint of Norwegian seafood products (Ziegler et al. 2003; Ziegler and Valentinsson 2008), and energy use in global salmon farming (Ayer and Tyedmers (2009); Nijdam et al. (2012)). Likewise LCAs comparing different farming methods have also been published (e.g. Aubin et al. 2009; d'Orbcastel et al. 2009b; Jerbi et al. 2012). Aquaculture, as a food production system, involves: diverse and multidisciplinary aspects, interlinkages amongst them, and highly variable production processes (e.g. different species and farming requirements, diverse production systems, and locations). This, coupled with the lack of transparency of the industry (Badiola et al. 2012), which makes difficult obtaining reliable data to represent all year around conditions, ends with an exhaustive data inventory and hinders a realistic comparison between studies. This complexity has limited the usability of traditional LCA methodologies (e.g.Wegener et al. 1996; Ellingsen and Aanondsen 2006; Finnveden et al. 2009; Samuel-Fitwi et al. 2012b). In this context, the authors reviewed the most significant publications in food production to assess the usefulness of LCA for aquaculture. As a result, a SWOT analysis was undertaken (conclusions shown in the supplementary material). One of the threats, presented as an outcome in the analysis and already mentioned before, was the complexity of aquaculture, limiting results comparison among studies; and this being directly linked with the lack of transparency for data collection in the industry. Consequently, LCAs are often based on generic and average data given by a database (i.e. no system-specific data), which leads considering diverse assumptions and obtaining so, wrong conclusions. In contrast, the multi-criteria approach of the LCA and the possibility of identifying critical points of processes can provide the framework to support the weaknesses mentioned. Some of the specific limitations detected in the aforementioned literature review have been solved in the past by combining different methods, such as LCA with Ecological Footprint (Samuel-Fitwi et al. 2012b), energy analysis with greenhouse gas emissions (Colt et al. 2008), LCA with Emergy Accounting (Wilfart et al. 2013), and the combination of LCA with Data Development Analysis (Ramos et al. 2014). Even so, the need for a broader range of science-based decision-making tools for aquaculture has been highlighted (e.g. Samuel-Fitwi et al. 2012b).

In aquaculture, and particularly in RAS, energy consumption is dependent on several factors such as species, rearing water temperature, climate and system configuration/design or layout and management. Furthermore, onsite energy consumption follows a time-based pattern (Ioakeimidis et al. 2013). Cumulative Energy Demand has been commonly used in environmental assessment method, such as Life Cycle Assessments (LCAs), as a single indicator of energy consumption (Frischknecht et al. 2015) when calculating different energy demands of the studied systems. Hence, limiting energy to a single value (e.g. an average value for a product or process) as resulted in the Cumulative Energy Demand indicator, may not reflect the reality of the farm, and energy saving measures cannot be accurately proposed. Energy audits provide an adequate proceeding/scheme through a detailed recording of energy flows. They provide real data (i.e. system-specific data) and estimate the energy consumption of a given system or process throughout a given period defining time-based energy-saving measures from both economic (€) and environmental terms (for example, with respect to CO_2 eq. emission). Consequently, an energy audit can proffer the energy model of a production cycle, by showing the energy consumption pattern of each of the devices forming the system. Thus, they may procure the best framework to quantify on-farm time-based energy consumption and in this manner provide more reliable and real data to be included in the LCA's data collection procedure. LCA in the seafood sector is fairly new compared to the development of this method in other sectors, such as petrochemical industry (e.g. Neelis et al. 2008), food and beverages (e.g. Ogunjuyigbe et al. 2015), and industrial in general (Boharba et al. 2016). Energy audits have also aided to reduce fuel and electricity costs and to increase predictable earnings in the fishing sector (Basurko et al. 2013), , especially in times of high energy price volatility; but it is not widespread activity. However, their inclusion as part of the life cycle inventory within the LCA has not been widely used but yet recommended (Nisbet et al. 2002). In contrast, in aquaculture and, particularly in RAS, among more than 20 LCA and system energy consumption related works published (Colt et al. 2008; d'Orbcastel et al. 2009b; Eding et al. 2009; Buck 2012) only one regards to energy efficiency (Ioakeimidis et al. 2013).

The contribution presented herein proposes a combined methodology (LCA with energy audits), which objective is to increase the precision of LCA results. The audits permit more accurate and system-specific data to be included in the Life Cycle Inventory (LCI) of the LCA by using detailed system's energy consumption quantification, temporally and spatially representative, that the data provided by the standard Cumulative Energy Demand indicator. Thus, this will help making a more precise diagnosis of the studied systems (i.e. already existing as well as new systems) and a possible energy consuming map. LCA studies reveal emission hotspots along the whole product value chain allowing to identify opportunities for improvements. Its combination with energy audits may offer an opportunity to substantially improve the assessment and the efficiency of the systems, by giving additional to use in the assessment. This will ultimately enable the proposal of time-based eco-design measures, which will depend on seasonality and particular conditions of the sea.

The methodology is implemented to assess the sustainability of a marketable size cod (*Gadus morhua*) production pilot-scale RAS facility located in the Basque Country. This species is one of the most important in the Basque households; the current consumption being 3,500 T/year while salmon consumption (the fastest growing species among the most popular species for the Basque consumers) is 2,800 T/year (MAGRAMA 2014). Nowadays, the principal on-growing method for the codfish aquaculture is through marine net pens (Bjornsson and Olafsdottir 2006) - only Fülberth et al. 2009 reported an attempt of on-growing codfish to marketable size utilizing RAS. This particular situation makes also difficult to obtain reliable data for the study. Species such as salmon which is currently reared in RAS (e.g. Summerfelt et al. 2013), presents a wider optimal rearing temperature range, which facilitates the rearing conditions by making the water temperature a lesser problem (e.g. more or less energy used to cool down the water, varying energy costs). Hence, the application of the combined methodology to a pilot-scale RAS production operation may better aid in identifying the main environmental hotspots of the systems and consequently, may assist to define a viable impact assessment methodology for the RAS industry making it more cost-effective.

2. The proposed methodology (Energy Audit + LCA):

2.1. Proposed methodology

In the proposed methodology the data obtained from energy audits are fed to the LCI step of a traditional LCA to provide additional information regarding energy consumption (Fig. 4.1). As a result, the additional data may give more insight to the energy consumption patterns of the production system, making possible periodical analysis and thus improve the environmental performance of RAS units.

Each of the methodologies is differentiated by different box and the proposed methodology is divided into 6 different working levels/steps:

- STEP 1, called goal and scope definition as included in traditional LCAs, defines the objectives and limits of the study. Here the design of the system should be described. A RAS can be differently constructed (e.g. diverse working devices can be employed, production factors are variable), which directly determines created impacts and consumed energy.

- STEP 2 is the LCI, where data considered for the study is compiled. Data include: foreground data (i.e. specific and relevant data for each production) and background data (i.e. data available in databases or literature). Moreover, this step integrates the results of the energy audit methodology into the LCI. This will aid to know the real energy consumption and to identify and characterize the factors affecting it (AENOR 2010). The energy audit includes the following procedure: (I) identification of energy consuming devices and factors affecting consumption fluctuations; (II) installation of energy measuring equipment; (III) energy monitoring; (IV) data collection, assessment and analysis; and (V) diagnosis. This step gives the information related to system's energy consumption and its pattern.

- STEP 3 is the Life Cycle Inventory Assessment (LCIA), assisted at the same time by different sensitivity analysis to assess which of the hypothetical scenarios studied are the most appropriate to implement.

- STEP 4 is the identification of environmental hotspots and saving measures. These measures are aimed to reduce both environmental impacts and economic costs presented by the system.

- STEP 5, is the economic feasibility, which gives the return period of the proposed measures. During the whole assessment and at different levels, diverse data/result interpretations are also proposed.

- Finally, STEP 6, called eco-designing and which is achieved improving the energy efficiency of products/systems, reducing energy and resource consumption.

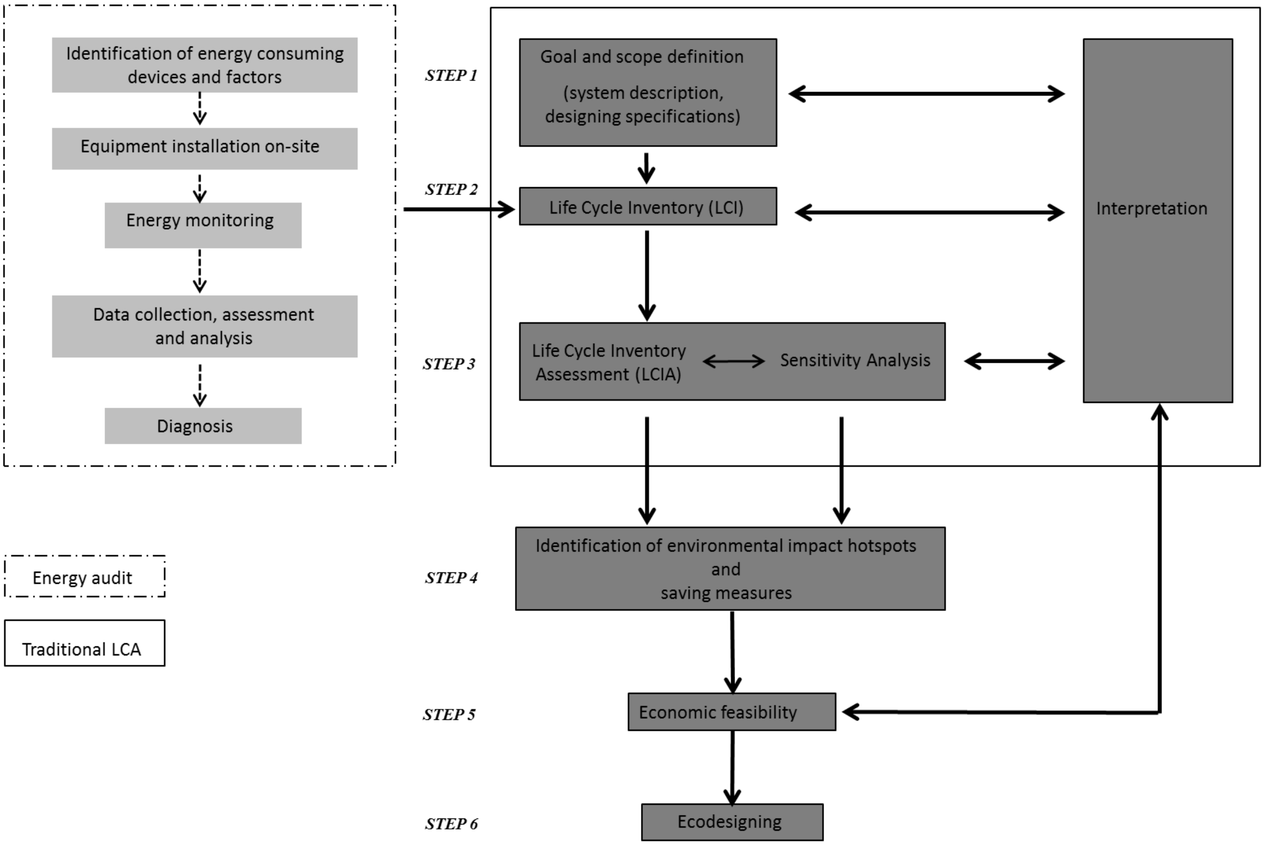


Fig. 4.1 Proposed methodology: integration of energy audits in the LCA methodology.

2.2. Case study

2.2.1. Description of the system

The proposed methodology was applied in the experiment of rearing Atlantic cod (*Gauds morhua*) in the pilot-scale RAS described in the “experimental design” section.

2.2.2. Application of the proposed methodology

STEP 1: Goal and scope

The Functional Unit (FU) was 1 kg of grown out cod, before slaughtering. Four inputs were considered in the analysis: the feed, the juvenile fish, the oxygen and the energy required to run the RAS unit. Thus, the analysis covered cod production between the arrival of the juvenile fish and raw materials, through to harvest of the market-size fish, including the transportations. Fig. 4.2 shows the scope of the assessment.

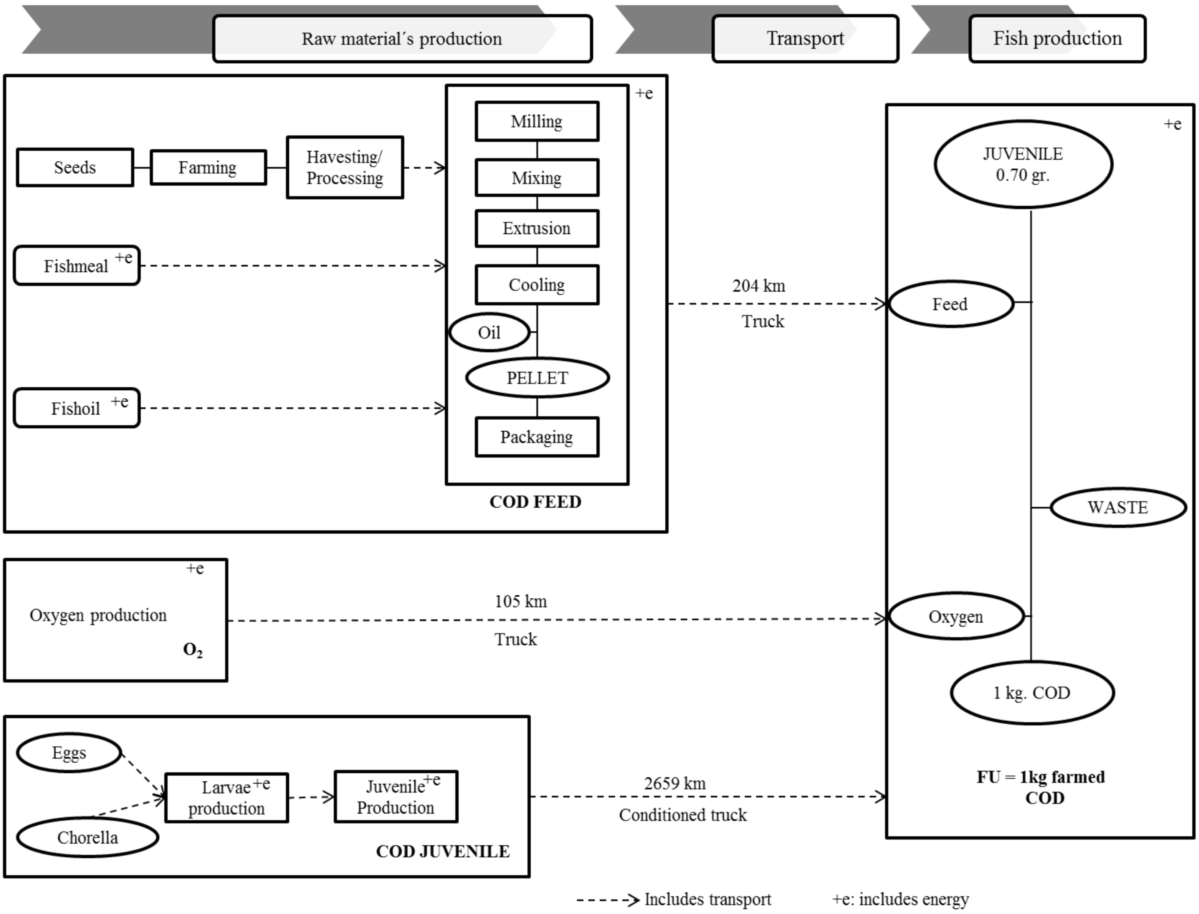


Fig. 4.2 Scope: main system and subsystems taken into account in the LCA study.

STEP 2: Life Cycle Inventory (LCI)

Data were obtained from a variety of different sources. Foreground data considered included: feed ingredients, juvenile fish production, oxygen quantity and transport, and energy consumption (monitored and registered continuously by the energy meter embedded in the RAS unit and measured by a portable electric power logger during a given period).

Feed was provided by a company located 204 kilometres southwest from the farm. Feed data, provided by the manufacturer, included: (I) ingredients origin, inclusion levels, transportation distances to the manufacturer and energy used in the production; and (II) feed transportation distances to the RAS unit and quantity required in the FU's production. Trawling's fuel consumption (65% of the feed used in the present study comes from the fisheries: 55% fish meal + 10% fish oil. Table 4.1), transportation of fish to slaughterhouse and transformation of wild fish into fishmeal and fish oil (i.e. energy required), were also included according to data in the selected database. At the same time, the allocation of environmental burdens between co-products, if any, was performed according the economic value of the co-products (i.e. production of feed ingredients).

Cod juveniles (average of 70.9 ± 20.34 g) were provided by a Norwegian company called *Fosen Aquasenter* (Trondheim) and they were transported in a conditioned (i.e. oxygenated, temperature-controlled) truck. The energy consumed by this transport was incorporated into the analysis. Egg to juvenile stage was also included in the analysis (including feed, transport, rearing conditions and energy needed) and data were obtained from both juvenile fish producers and literature sources. Additionally, in aquaculture, some species require microalgae as nutritional supplementation in their larval life stages. In this assessment, production of chlorella, the micro-algae species used in cod larval nutrition, was included. The unique nature of this dietary requirement makes its inclusion necessary within the assessment; its quantity and transport were incorporated. Waste produced in the on-growing stage and included in the analysis included wastewater, uneaten feed and faeces.

Oxygen was provided by a company with several locations within the surrounding area; thus an average mileage was calculated for transport purposes. The capacity utilization of the trucks, in both feed and oxygen transportation, was considered as full trucks delivery. Energy required for oxygen production was taken from the bibliography (Sonani and Ratnadhariya 2013) and the way of transport and volume required were also included. The oxygen required by the fish was calculated according to the species biological requirements.

The energy audit followed the procedure presented in Fig. 4.1. For this particular case study energy consuming devices were: the heat pump, the main and secondary pumps, the skimmer and UV systems and the factors affecting consumption fluctuations were: temperature, flow rate variations and system maintenance. The equipment used for the energy measurement was: a fixed energy meter and a portable electric power logger (FLUKE 435 Series II, power quality and energy analyser, *by FLUKE*). The first one continuously registered the energy consumed by the RAS unit along the whole experiment providing an absolute value (i.e. kWh/period). The second one, i.e. portable energy meter, registered each of the devices during a week, and thereafter, an extrapolation was made according to the devices working hours for the whole experiment. Hence, this number may differ with the continuously monitored data. Furthermore, not all the devices were operating during the whole experiment; the heat pump worked when the rearing temperature exceeded experiment´s limit and the rest of the devices operated continuously, unless maintenance operations were carried out. Data obtained from the energy audit were included in the LCI, giving additional information about the energy consuming pattern along the experiment.

Table 4.1. LCA inventory: quantity, origin, transport mode, distance to farm and references (Functional Unit: 1 kg of cod)

INPUTS – Subsystems production	Ingredients		%	Origin	Transport mode	Distance (km)	Data source (reference)
Feed	Fish meal[1]		55	Peru	Container ship	9,130	Manufacturer
	Gluten		14	UK	Truck-bulk	1,480	Manufacturer
	Fish oil [2]		10	Peru-Chile	Container ship	10,900	Manufacturer
	Wheat		5*	Spain	Truck-bulk	400	Manufacturer
	Concentrated soybean protein		5*	France	Truck – Bulk	800	Manufacturer
	Pea meal		5*	France	Truck – Bulk	800	Manufacturer
	Corn gluten		5*	France	Truck – Bulk	800	Manufacturer
	Vitamins[3]	A	5000				
		E	150				
	Oligoelements[3]	E4	40				
		E2	2				
		Zn	90				
		Mn	15				
	Feed production	460.00	kWh/kg				Boissy et al. 2011
Juvenile fish	Hatching and nursing[4]	14.84	kWh	Norway	Conditioned truck[5]	2,639	Producer
	Growing phase[4]	3.15	kWh				Producer
Oxygen		0.19	kWh				Sonani and Ratnadhariya 2013
INPUTS – On-growing							
Oxygen		0.21	kg	Spain	Truck	100	Timmons and Ebeling 2010
Juvenile fish		0.07	kg				Present contribution
Energy for farming[6]	On-growing	29.40	kWh				Present contribution
Feed quantity (FCR) [7]		1.57	kg				
Feed transportation to farm					Truck	204	Present contribution

Key:
*estimated average values
[1] obtained for anchovy (fishing fuel and energy for production included).
[2] obtained for both farmed and wild salmon and tuna (fishing fuel and energy for production included).
[3] these values are given in mg/kg.
[4] obtained from Iribarren et al. 2012.
[5] includes 1.098 kg of refrigerant R-22.
[6] obtained from an energy meter, average value for the whole experiment
[7] Food Conversion Ratio: mass of feed bought per mass of fish sold (losses in the production accounted.) Calculated as an average of three tanks.

Finally, the background data considered included: (I) raw materials processing; and (II) oxygen production. These data were taken from the EcoInvent 3.0 database. The LCI is summarized in Table 4.1.

STEP 3: Life Cycle Impact Assessment

The impact assessment was in accordance with the ISO 14044 guidelines and impact categories were selected based on the relevance to reflect the environmental impacts of RAS and literature review. At a global scale: abiotic depletion (AD), global warming potential (GWP) and energy use were the principal categories, whereas at a regional scale eutrophication potential (EP) and acidification potential (AP) were the primary categories. The sensitivity analysis was made based in two variables: (I) energy consuming pattern, differentiated by the time of the year and; (II) the energy source (i.e. energy based on fossil fuels and/or on a renewable source). One of the objectives of an energy audit, stated by AENOR 2010, is to detect and evaluate different energy saving and diversifying opportunities. Thus, the possibility of using renewable energies was assessed. The renewable source was biogas obtained from agricultural plants.

Three different scenarios were assessed to see the effect of the energy consumption fluctuation in the LCA results: Scenario 1 (SC. 1) considered the average energy consumption value of the whole experiment; Scenario 2 (SC. 2) included the energy consumption value corresponding to the time of the year where the energy consumption was minimum, and Scenario 3 (SC. 3) to the maximum consumption value. All scenarios included an additional dual option regarding the source of energy: option A, where 100% of the energy source was non-renewable (NR) and option B, where 50% of the energy was NR and 50% was renewable (R).

These scenarios were decided aided by the results obtained in the energy audit, which provided an energy consuming pattern during the whole experiment showing at the same time the main energy consuming device.

Finally, *STEP 4, 5 and 6* (i.e. *identification of environmental impact hotspots and saving measures, economic feasibility and eco-design*) were proposed steps to be followed in the combination of both (i.e. LCA and energy audits) methodologies. Although their accurate study was beyond the present contribution, feasible different energy sources and saving measures are given.

The software SimaPro 8.0.2® and CML Baseline 2000 v 2.0 impact assessment method were used to do the calculations (Heijungs et al. 1992; Guinée et al. 2001).

3. Results

Tank’s water temperature maintenance consumed the largest percentage of energy (Table 4.2). More than half of the total energy consumed (56,114 kWh) was due to the heat pump (40,320 kWh). This outcome diagnoses the relevance of the rearing temperature in the total energy consumption (and thus in the costs). Energy consumption fluctuated along the experiment duration (Fig. 4.3), and it was mainly due to the heat pump´s daily energy consumption fluctuation (Fig. 4.4). Temperature and heat pump´s energy use are positively correlated (r = 0.86). Consequently, 2 different situations were identified: (I) when the heat pump was switched off the energy consumption reached its minimum, with an average value of 18.43 kWh/kg; and (II) when the heat pump was switched on the energy consumption reached maximum levels, with an average value of 40.57 kWh/kg.

Table 4.2. Energy consumed per each of the energy-consuming devices measured by the power quality and energy analyzer.

Pilot-scale devices	*kW*	*kWh*[1]	*kWh/kg*	*Consumption (%)*
Main pump (25 Hz)	0.5	3,905.3	2.3	7.0
Secondary pump (25 Hz)	0.5	3,905.3	2.3	7.0
Skimmer	0.8	6,652.8	3.9	11.9
Ultraviolet1	0.1	695.5	0.4	1.2
Ultraviolet2	0.1	635.0	0.4	1.1
Heat Pump	4.0	40,320.0	23.7	71.8
Total		56,114.0	33.0	

Key:

[1] Time frame of 15 months, i.e. experiment’s period

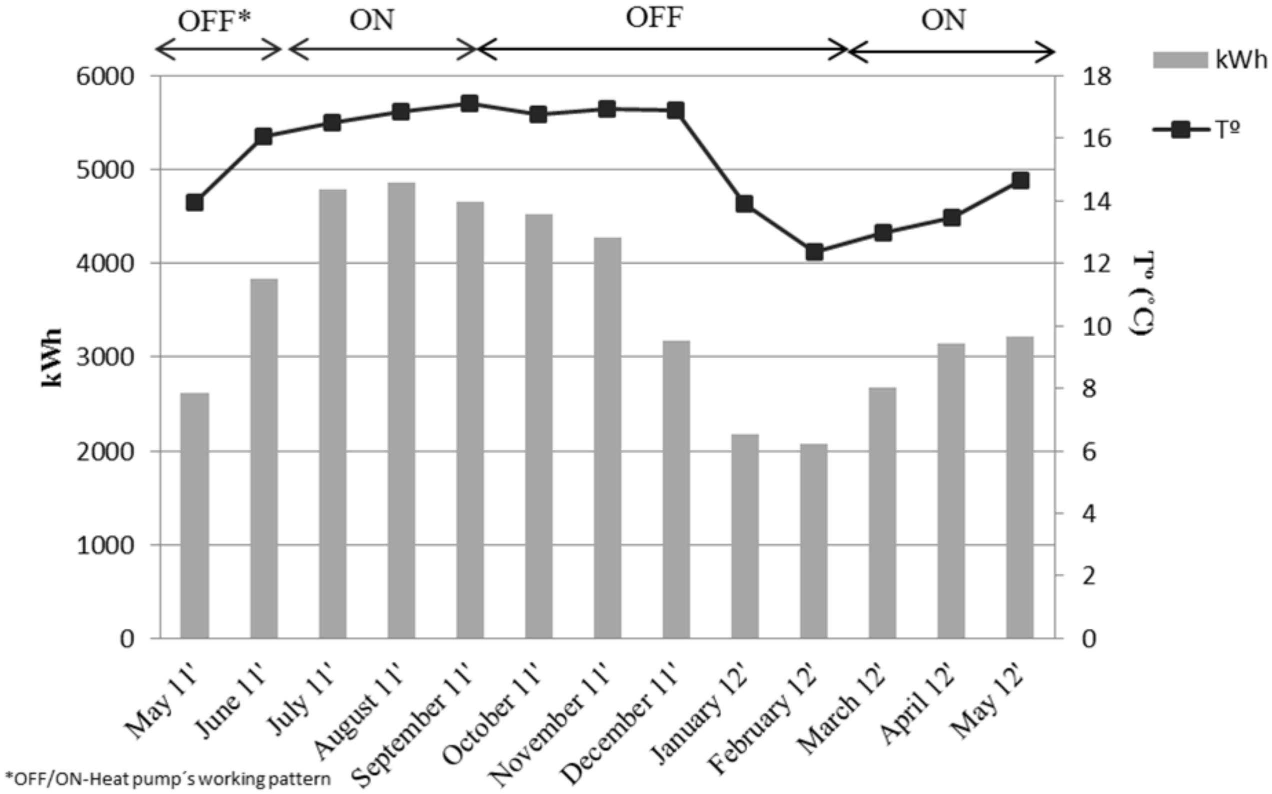


Figure 4.3 Heat pump´s working pattern according to RAS unit's water temperature, and its impact on the kWh consumption (May 2011- May 2012).

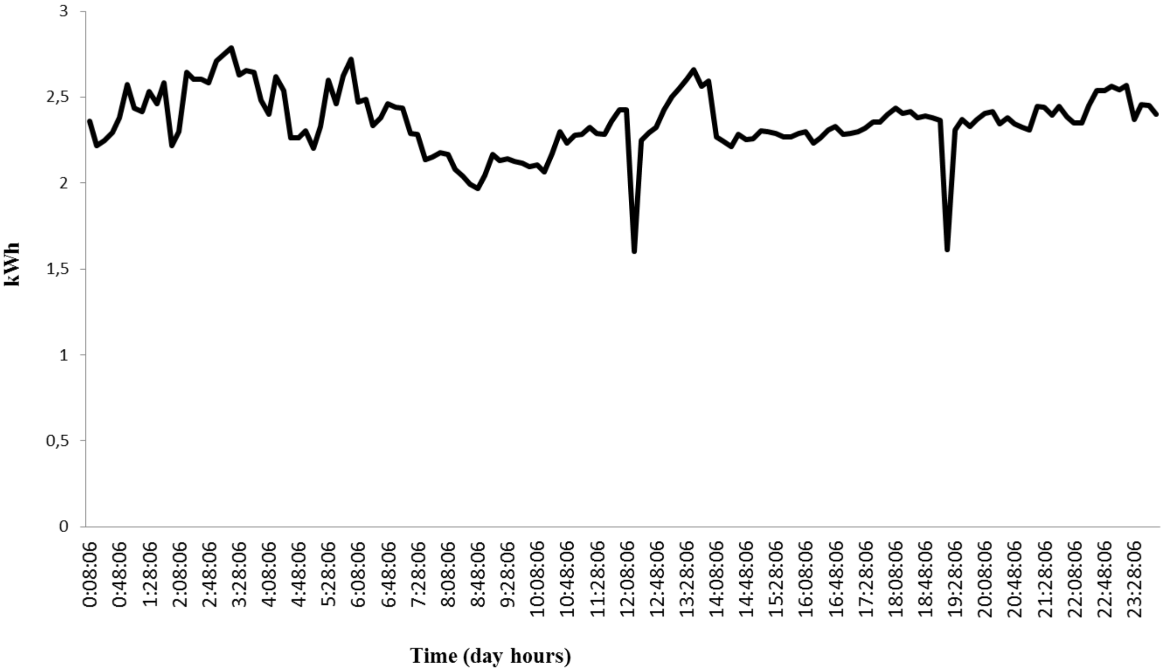


Figure 4.4 Example of the Heat pump's energy consumption during a specific timeframe i.e. one day (March 2012).

Results of the scenarios' analysis are listed in Table 4.3. Results represent life cycle impacts associated with the on-growing 1 kg of Atlantic cod in the Basque coastal area. Out of all the impact categories assessed (i.e. AD, AP, EP and GWP), the main environmental impact contributor in SC.1 option A (i.e. average energy consumption using 100% non-renewable energy) was the on-farm electricity use (Fig. 4.6), representing nearly 80 % of the total environmental footprint. At the same time, in 3 of the impact categories (i.e. AP, EP and GWP) the pattern was similar, where oxygen, juveniles and feed, in this order, were the less impactor. Results were similar for the option A in both SC. 2 and SC. 3 (i.e. oxygen, juvenile fish and feed, in this order, were the less impactor in AP, EP and GWP impact categories while consumed energy was the main contributor in all four). Even though there was an evident energy consumption fluctuation (i.e. energy consumed in SC. 3 doubles SC. 2), the main impact contributor was the energy. At the same time, in SC.2, where energy consumption was less than the half of the maximum of SC.3, the overall impacts were decreased although in the same proportion (e.g. in SC. 2 GWP was 58.00 kg CO_2 eq / kg fish while in SC 3. 75.33 kg CO_2 eq / kg). Additionally, the results differences between SC. 2 and SC. 3 due to the energy consumption had similar impacts in the rest of the inputs (i.e. juvenile fish, oxygen and feed); differences varying between 40-45 %.

Equally, for the scenarios where 50% of the consumed energy was from a non-renewable source and the other 50% renewable (i.e. biogas obtained from agricultural plants), oxygen, juveniles and feed were the less impactors in AP, EP and GWP impact categories. Here, analyzing energy as a single input, renewable energy source represented 6 % of the impact and the remaining 94 % came from non-renewable source. At the same time, impacts created by the energy were severely decreased from option A (i.e. 100 % NR) to B (i.e. sum of 50 % NR and 50 % R) in 3 of the scenarios, and this reduction was mirrored in feed, juvenile and oxygen (in this order), where part of the inputs are related to the energy consumption, i.e. non-renewable energy source. Although the inclusion of renewable energy seems to be insignificant, it considerably decreases the overall impacts created by the RAS unit (Table 4.4), representing up to 50 % in some of the cases (i.e. EP in SC. 3).

Table 4.3. Life cycle impacts associated with the on-growing of 1 kg of Atlantic cod in the Basque coastal area during 15 months (FCR 1.57). Scenario 1 (SC. 1) corresponds to the average energy consumption (29.40 kWh/kg) during the whole experiment; current situation of the RAS unit. Scenario 2 (SC. 2) corresponds to the minimum energy consumption (18.43 kWh/kg). Scenario 3 (SC. 3) corresponds to the maximum energy consumption (40.57 kWh/kg).

		Juvenile fish		Feed		Oxygen		Energy		
	Impact category	SC. A[2]	SC. B[3]	SC. A	SC. B	SC. A	SC. B	SC. A	SC. B	
		100% NR[4]	50/50 - NR/R[5]	100% NR	50/50 - NR/R	100% NR	50/50 - NR/R	100% NR	50% NR	50% R
					SC. 1					
AD	%	12.36	19.62	10.64	17.32	0.24	0.39	76.80	0.82	62.31
	kg Sb eq	0.02	0.02	0.02	0.02	3.44E-04	3.44E-04	0.11	7.3E-04	0.06
AP	%	6.57	10.34	12.64	19.78	0.25	0.39	80.62	5.43	64.22
	kg SO_2 eq	0.01	0.02	0.02	0.02	4.41E-4	4.41E-04	0.14	6.07E-03	0.07
EP	%	7.07	11.32	11.83	18.34	0.15	0.23	81.21	6.07	64.37
	kg PO_4^{-3}	2.33E-03	2.32E-03	3.79E-3	3.79E-03	4.81E-4	4.81E-05	0.03	1.26E-03	0.01
GWP	%	12.15	17.77	18.93	27.72	0.25	0.37	68.81	2.98	51.35

	kg CO_2 eq	2.61	2.61	4.08	4.08	0.05	0.05	14.92	0.44	7.56
SC. 2										
AD	%	17.22	25.52	14.91	22.12	0.34	0.51	67.52	0.68	51.38
	kg Sb eq	0.02	0.02	0.02	0.02	3.44E-04	3.44E-04	0.07	4.63E-04	0.03
AP	%	9.39	13.83	18.46	26.56	0.36	0.53	72.23	4.62	54.67
	kg SO_2 eq	0.01	0.01	0.02	0.02	4.41E-04	4.41E-04	0.09	3.84E-03	0.05
EP	%	10.12	14.94	16.92	34.68	0.21	0.31	72.84	5.18	54.95
	kg PO_4^{-3}	2.34E-03	2.28E-03	3.83E-03	3.79E-03	4.81E-05	4.81E-05	1.64E-03	7.94E-04	8.42E-03
GWP	%	16.33	22.12	25.42	55.32	0.34	0.46	58.00	2.35	40.94
	kg CO_2 eq	2.61	2.61	4.08	4.08	0.06	0.05	9.33	0.28	4.79
SC. 3										
AD	%	9.54	15.85	8.24	13.73	0.19	0.31	82.14	0.92	69.34
	kg Sb eq	0.02	0.02	0.02	0.02	3.44E-04	3.44E-04	0.15	1.00E-03	0.08
AP	%	5.03	8.12	9.65	15.67	0.19	0.31	85.12	5.93	70.12
	kg SO_2 eq	0.01	0.01	0.02	0.02	4.41E-04	4.41E-04	0.19	8.38E-03	0.10
EP	%	5.41	8.74	8.99	14.56	0.11	0.18	85.55	6.64	70.12
	kg PO_4^{-3}	2.33E-03	2.32E-03	3.79E-03	3.79E-03	4.81E-05	4.81E-05	0.04	1.73E-03	0.02

GWP	%	9.58	14.78	15.00	22.93	0.20	0.31	75.33	3.42	58.73
	kg CO_2 eq	2.61	2.61	4.08	4.08	0.05	0.05	20.52	0.61	10.44

Key: [1] AD: Abiotic Depletion; AP: Acidification Potential; EP: Eutrophication Potential; GWP: Global Warming Potential

[2] A: 100% of the energy comes from a non-renewable source
[3] B: 50% of the energy is from a non-renewable source and 50% from a renewable source
[4] NR: non-renewable energy (Spanish grid medium voltage: 1 kWh = AD 1.04E-03 kg Sb eq; AP 1.36E-03 kg SO_2 eq; EP 2.51E-04 kg PO_4^{-3}; GWP 0.143 kg CO_2 eq)

[5] R: renewable energy (i.e. biogas obtained from agricultural plants: 1 kWh = AD 9.4E-06 kg Sb eq; AP 1.11E-04 kg SO_2 eq; EP 2.25E-05 kg PO_4^{-3}; GWP 7.64E-03 kg CO_2 eq)

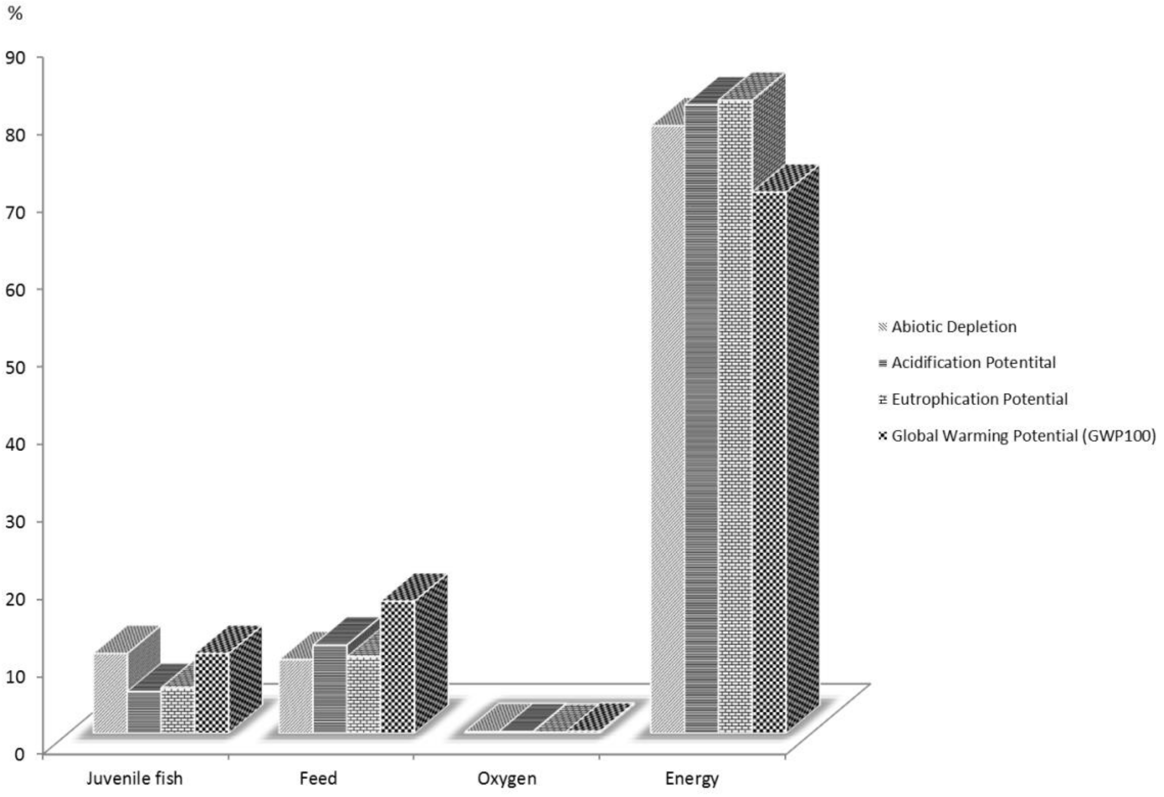


Fig. 4.5. Environmental footprint of 4 of the subsystems considered in the LCA for SC. 1, with 100% of non-renewable energy sources (i.e. current situation of the RAS unit).

The sensitivity analysis made to compare the total energy consumption in different periods (i.e. S.C. 1, S.C. 2 and S.C. 3) showed that the main contributor input was the energy consumption in all the impact categories assessed. This outcome remarks the importance of the energy consumption within the RAS unit and suggests that both environmental impacts created and costs will be reduced when decreasing the amount of system's energy use and the type of energy use. Furthermore, the inclusion of data obtained from the energy audit aided to know which factor (i.e. heat pump use) affects the main impact contributor input (i.e. energy consumption), and thus, propose different options (i.e. A and B) to analyse; creating system specific scenarios and seeking possible impact reduction. As resulted from Table 4.4, the differences between the impacts created in different periods (i.e. S.C 2 and S.C. 3) were substantial and the use of renewables in such periods modifies the impact created considerably. Such time-based information was obtained including the results from the energy audit in the LCA methodology.

Table 4.4. Summation of the impacts created in options A (i.e. 100% of the energy source was non-renewable) and B (50% of the energy was non-renewable and 50% was renewable).

Impact category		A (100 % NR)	B (50/50 % NR/R)
		SC. 1	
AD	kg Sb eq	0.15	0.10
AP	kg SO_2 eq	0.17	0.12
EP	kg PO_4^{-3}	0.04	0.02
GWP	kg CO_2 eq	21.64	14.74
		SC. 2	
AD	kg Sb eq	0.11	0.07
AP	kg SO_2 eq	0.12	0.08
EP	kg PO_4^{-3}	0.01	0.02
GWP	kg CO_2 eq	16.08	11.81
		SC. 3	
AD	kg Sb eq	0.19	0.12
AP	kg SO_2 eq	0.22	0.14
EP	kg PO_4^{-3}	0.05	0.03
GWP	kg CO_2 eq	27.24	17.75

4. Discussion

RAS are a sustainable way of producing fish (Martins et al. 2010). Environmental impacts such as Eutrophication Potential, which are very dependent on the exact time and place occurred, decreases in comparison with other production systems by collecting and treating wastewater and even sequestering some waste nutrients for reuse (Colt et al. 2008; Ayer and Tyedmers 2009; d'Orbcastel et al. 2009c). In contrast, impact directly linked to energy use, including Abiotic Depletion (i.e. the depletion of non-renewable resources), Global Warming Potential and Acidification Potential, are substantially higher (Ayer and Tyedmers 2009). In this particular case, on-growing one kg of cod resulted in the release of around 22 kg of CO_2 equivalents to the atmosphere (Table 4), compared to just over 2 kg CO_2 equivalents in the net-pen system studied by Ayer and Tyedmers (2009). One of the reasons for such difference may be due to larger transportations (i.e. increase in kg CO_2 eq produced) required for the inputs included in the analysis (i.e. feed ingredients were from South America and countries around Europe and juvenile fish from Norway). In fact, some has stated feed production as the issue of concern in the industry (Aubin et al. 2009; Ellingsen et al. 2009; Jerbi et al. 2012; Pelletier and Tyedmers 2007), basically due to impacts created by the trawling stage (i.e. fuel consumed); while others, in accordance with the present study, highlight the energy usage during the rearing of the fish as the main impact producer (Aubin et al. 2006; Colt et al. 2008). This is in accordance with the present study, where the energy consumption was mainly due to the heat pump's work. Generally, energy's environmental impact comes from the sum of feed production (including fishing stage) and electrical/fuel energy required for rearing; they count 91-99% of the total energy consumption (Colt et al. 2008). In fact, d'Orbcastel et al. (2009c) allocated to system's operation 70 % of the total energy consumption. Therefore, energy use in general (i.e. during different stages of the production including feed production and on-farm energy consumption) is of great importance in RAS.

Energy used by the studied RAS unit (i.e. 29.40 kWh/kg of energy consumption on average) had the largest impact in four of the categories and in 3 of scenarios analysed. Because of the use of large amounts of fossil energy sources, Acidification Potential (0.14 kg SO_2 /kg fish) and Global Warming Potential (14.92 kg CO_2 eq) reached high levels. This result are similar to the ones obtained by Ayer and Tyedmers (2009). Moreover, results showed different values for the impact categories between scenarios where the oxygen, juvenile and feed inputs, in this order, were the lowest contributors. Regarding to the feed, values for the impact categories in this study are in the same range as in studies by Ayer and Tyedmers (2009). However, compared to our study, Ayer and Tyedmers (2009) found much lower values for Global Warming Potential (-65%). This may be explained by the transportation of the feed to our location (i.e. kg feed / km of transport) which was made on demand and in low quantities (i.e. pilot-scale production and not commercial scale). The lack of contributions regarding the impacts created by the use of renewable energy in RAS in terms of e.g. Acidification Potential and Global Warming Potential make difficult its comparison. According to the database used in the present contribution, non-renewable energy production presented higher environmental impacts in four of the categories assessed when comparing to renewable energy production (e.g. 6.74 E-02 and 1.24 E-03 kg CO_2 eq, respectively). This may explain the low impact values generated by the renewable energy used (i.e. biogas from agricultural plants) although the same amount of energy was used (i.e. 50/50 %). Overall, the scenarios comparison reflects the variation of the impact associated with the production of cod during a given timeframe, showing the importance of having time-based information throughout the production cycle.

In relation to the energy use, values in the literature range from 17 to 23 kWh/kg fish: 17.55 kWh/kg of salmon produced (Ayer and Tyedmers 2009), 20.04 kWh/kg of turbot produced (Iribarren et al. 2012) and 22.6 kWh/kg of trout produced (d'Orbcastel et al. 2009b). The energy audit here presented concluded that the energy use varied considerably from 18.43 to 40.57 kWh/kg fish between different periods (i.e. heat pump use); hence, this confirms the importance of the energy quantification along the production cycle (i.e. not relying on an average value) in order to know when the energy saving measures should be applied with effective results. Nevertheless, the maximum value obtained during the heat pump's use (i.e. 40.57 kWh/kg fish) did substantially differ from the literature which could be possibly explained by different reasons: (I) operational and designing factors such as farm location, system's layout (i.e. head losses), species produced (i.e. water temperature required) and the rearing stage and; (II) the possible oversizing of devices and a non-proper management (i.e. non-qualified people in charge of the system) (Badiola et al. 2012). As mentioned, temperature maintenance's, depending on the species, would lead more or less the energy consumption. In this particular case, the energy consumption is mainly due to the local requirements on seawater temperature for correct maintenance and subsequent optimization of cod fish growth performance and production. The experiment's maximum allowed temperature was set up at 16.0 ± 1.5ºC, according to cod´s maximum tolerated growth temperature (Bjornsson et al. 2001). Basque coastal water temperature profile can vary from 12ºC to 23ºC during a year (monthly mean Sea Surface Temperature (SST) (Goikoetxea et al. 2009). At a local basis, this wide temperature range makes it necessary to chill the rearing water, a process that is extremely energy intensive. This operation takes places during summer periods (i.e. May-September), although it will clearly depend upon each year weather´s conditions. In this particular case, the heat pump´s functioning dictated RAS unit´s total energy consumption. This may suggest that the selection of appropriate devices, meaning properly designed cost-effective devices as well as suitable energy sources is pivotal for having a successful business (Badiola et al. 2012). An example of this is the profitable production of tilapia and shrimp on-growing in RAS which operates with large amount of water in an environment where water is scarce, such as in the Arizona desert. In this particular case water is naturally heated by the use of geothermal energy, making the production cost-effective (Buck 2012). Thus, it is assumable to be more efficient (i.e. less costly) to farm warm water fish in cold weather than it is to farm cold water fish in warm weathers. This comes from the possibility of: (I) having access to non or low-cost energy sources (such as geothermal or waste heat from industry, respectively) and; (II) not having to rely on fossil fuel source electricity. In contrast, the chilling of water requires the use of electricity, increasing both environmental and economic costs. A possible strategy could be running a flow-through production in winter (i.e. taking advantage of natural temperature) and closing to a RAS in summer, which would reduce unit´s power consumption. Furthermore, system design improvements such as using low-pressure filtration (i.e. drum filters and gravity-operated biofilters) instead of rapid sand filtration and pressurised biofilters used in this study, may lower RAS energy consumption (Timmons and Ebeling 2010). Moreover, measuring energy consumption along the production cycle through an energy audit and thus differentiating consumption peaks (i.e. maximum and minimum) could help in the design on an energy-efficiency plan (e.g. using renewable energy sources and contracting adequate energy rates in the maximum energy consumption periods). Thereafter, by using more energy-efficient systems (together with a proper business plan and designed system), species selection may be done based on market demands instead of prevailing environmental conditions as this will determine whether such demand can be met at expected market prices whilst keeping the business profitable.

Fossil fuels supply 80 % of the total energy demand worldwide; however, renewables are the fastest growing energy sources (a growth rate of 2.5 % per year) (EIA 2014). The high level use of non-renewable energy (i.e. large amounts of fossil fuels) indicates that the Acidification Potential and Global Warming Potential impact categories are much higher in RAS than in traditional flow-through systems (e.g. Aubin et al. 2006). Thus, although very few examples have been reported (Toner 2002; OPP 2015), renewable energies have the potential of being used in RAS as long as they are placed in suitable locations with access to energy sources such as solar; wave; hydro; thermo-solar; and domestic hot water. Particularly in very exposed coastal regions, as the one considered within the presented study, a potential solution to decrease non-renewable energy use could be wave energy. The coastal orography, wave currencies and their energy content have made the study of this energy a logical and complementary source. As a particular case, on the Basque continental shelf, Wave Energy Converters may supply from 37 % to 50 % of the electrical consumption of local households, avoiding the annual emission of 0.96 to 1.54 million tons of CO_2 into the atmosphere (Galparsoro et al. 2012). Another solution could be the use of the cogeneration technique with biomass, or other types of clean flues. Biomass energy represents the 17 % of the total energy generated in the region, i.e. 535 MW of the total 3100 MW and 64 % of total renewable energy use (EJ-GV 2011) and impacts created for the energy consumed in the RAS unit could be decreased up to 35 % in average in four of the impact categories assessed (Table 4.4).

From an economic standpoint, the cost of electricity (0.13 €/kWh) (Iberdrola 2015) is one of the main constraints in Europe; this high cost hampers the promotion and development of new fish farming businesses. In this particular case, it may be technically and socially feasible to produce cod in the Basque Country (Badiola et al. 2016); however, this activity would be economically restricting. The electricity consumption for the rearing alone represents a production cost of 3.24 €/kg of cod, 40% of the total costs (Badiola et al. 2017). Therefore, this would affect the viability of setting up a RAS cod industry in this region and/or lead to proposed complex financial engineering considerations i.e., economies of scale. This context, and remarking the endorsement (i.e. funding 25% of the installation costs) of using renewable energies (especially biomass as a local resource) by the local government and institutions, may offer an opportunity for an environmentally sustainable and less costly eco-designed industry. On the other hand, the use of wind/or wave energy as renewable source which have been studied for their use in RAS. The installation of these stations may require a large capital outlay although it could be recouped within a period of 6 years (Toner 2002).

This study has corroborated the high energy demand of RAS. Thus, it seems obvious that there is a need for an energy efficiency plan, which should include several renewable energy alternatives and energy saving measures. Good practices, both in the design and management of the systems would aid in: (I) a more energy efficient framework; (II) reducing energy losses; and (III) adapting already existing systems to each particular production (Badiola et al. 2012; OPP 2014). Although there are no examples available in the literature or public databases regarding their use in aquaculture, frequency controllers, which are used to change the frequency and magnitude of the constant grid voltage to a variable load voltage, are shown to aid in the reduction of the electricity consumption of pumps; their efficiency is mainly dependent on the number of starts-stops and required water flows. Thus, according to the already mentioned successful examples, an average of 20% of the consumed energy could be saved in such a way. Furthermore, the system´s engines, pumps and lighting configurations, as well as thermal equipment’s isolation, all contribute to good practices in RAS design, operation and management. Additionally, the employees’ ability to understand the workings of the system and respond to issues effectively is vital for efficient management and operation (Badiola et al. 2012; OPP 2014).

The present contribution helps identifying some of the elements that comprehend part of the environmental and economic sustainability of RAS. It gives the required steps to follow in terms of presenting a more comprehensive LCA, providing a detailed scan of the system and indicating methods to improve the environmental appraisal. In this manner, a methodology is presented that constitutes a more comprehensive approach than those published so far by others (Ioakeimidis et al. 2013). Data obtained in the energy audit is included in the LCI improving the quality of data and resulting in a more accurate and system-specific assessment. Therefore, more efficient RAS units can be designed combining both methods as: (I) a time-based energy consuming graph may give the information needed to know the energy consuming fluctuations and all together assist on an improved mapping of the production framework; (II) more precise (i.e. system-specific) and period-specific recommendation of saving measures could be made; (III) the information given by the identification of the parameters will provide extra information to draft an energy consuming-map which at the same time may aid to know where to invest and the return period of that investment. These will improve the energy efficiency of the RAS unit, reducing energy and resource consumption. RAS units are highly management-dependent (Badiola et al. 2012); therefore, making precise and on-time decisions is crucial for their efficient functioning.

5. Conclusions

Energy should be one of the main aspects under study for the environmental and economic sustainability assessment and development of RAS. This contribution presents a combined framework of two different environmental assessment tools, LCA and energy audits, in order to achieve more accurate and representative data to be used for decision-making. Moreover, with the aim of monitoring and making precise decisions by time-based fluctuations of the system (e.g. temperature’s rapid variation due to an unexpected weather change or system´s manipulation for maintenance purposes), it would be useful to develop an approach which would monitor on a continuous basis each of the energy-consuming devices. The implementation of an entire energy audit in such “high-technology” system would be a useful tool for already operating (i.e. optimising their production by implementing more precise energy saving measures) and future systems. A detailed knowledge of the system´s performance would help to improve RAS´ efficiency and to expedite the usually complicated decision-making process.

Supplementary material: The use of LCA as a method to assess the environmental impacts of the aquaculture sector

	Statement	References[1]		Statement	References[1]
S	**Multi-criteria and multi-impact method for comparison across categories.** LCA gives a broader understanding of the environmental performance of products and it helps to identify trade-offs between different impacts (e.g. products with low carbon footprint but a greater dependence on biotic resources).	5,22,23	W	**Lack of transparency during the data collection**. Difficulties when obtaining data from different industry parties.	13
	Multi-scale assessment. The selection of impact categories allows to define if impacts are at regional and/or at a global scale (e.g. the global warming potential is at a global scale while the eutrophication potential refers to a regional scale).	6,8,11,20		**Limited number of LCA studies for marine products.** Inventory data, databases and impacts categories are yet in early stages.	1-5,15,22,24
	Identification of critical points of processes. LCA helps to identify which of the steps on a process are/have to be considered as hotspots and thus, require a more accurate analysis.	7,12,17,19		**Omission of natural environmental inputs.** LCA of agricultural systems such as aquaculture does not consider natural environmental inputs such as solar energy, rain, wind.	5,13,24
	Covers wide range of environmental impacts. In LCAs considered impacts go from the ones related to e.g. the atmosphere, to fresh or marine water, land use.	20,21		**Limited impact categories.** Impact categories adopted are typical to LCA research in other sectors (i.e. broad-scale environmental impacts characteristic of human industrial activities)	10,16,18, 19
				Not considers resource depletion. It looks both at the environmental consequences of the emissions and the amount of resource used, but not their depletion.	9,10,13,16,19,24
O	**Environmental criteria are compulsory.** When developing a LCA from an aquaculture activity, the environmental side of the production is without exception required to be assessed in order to have a real vision.	22	T	**In aquaculture, there is a great diversity of species** (which implies very diverse environments and growing conditions), **limiting thus the comparison among obtained results.**	11
	Combination with other methodologies. The combination of LCA with other methodologies (such as Emergy Accounting and Data Development Analysis) will give the opportunity to overcome with certain weaknesses such as the complexity of optimising biophysical efficiency in aquaculture systems and implementing a timeline perspective in the environmental assessment of fishing systems.	14,22-25			

Key.

S: strength; W: weaknesses; O: opportunities; T: threats

[1] References are:

1-Wegener et al. 1996; 2-Audsley et al. 1997; 3-Ceuterick 1998; 4-Haas et al. 2000; 5-Papatryphon et al. 2004; 6-Ellingsen and Aanondsen 2006; 7-Gronroos et al. 2006; 8- Thrane 2006; 9- Ulgiati et al. 2006; 10-Pelletier et al. 2007; 11-Aubin et al. 2009; 12-Ayer and Tyedmers 2009; 13Pelletier et al. 2009; 14-d'Orbcastel et al. 2009b; 15-Winther et al. 2009; 16-Ziegler et al. 2011; 17-Iribarren et al. 2012; 18-Jerbi et al. 2012; 19-Nijdam et al. 2012; 20-Samuel-Fitwi et al. 2012b; 21-Samuel-Fitwi et al. 2012a; 22-Vázquez-Rowe et al. 2012; 23-Samuel-Fitwi et al. 2013; 24-Wilfart et al. 2013; 25-Ramos et al. 2014

Contribution 5

Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges

This Contribution has been published in:

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Summary

The main issues for Recirculating Aquaculture Systems (RAS) are analyzed, in order to lead to better solutions for future managers, identifying possible areas for improvements and future challenges for the industry. RAS-based production companies, researchers, system suppliers and consultants were interviewed separately, in order to gain an overall understanding of those systems and what developments could assist, in a positive way. Answers and subsequent analysis identified as significant barriers: poor participation by the producers; a disincentive on sharing information; and a lack of communication between different parties. The main issues are poor designs of the systems, as many had been modified after a previous approach was unsuitable; and their poor management, due mainly to an absence of skilled people taking responsibility for water quality and mechanical problems. As RAS will play an important role within the future of aquaculture, their enhancement is needed. Key priorities are the necessity to improve equipment performance, through researching at a commercial scale and further work on the best combinations of devices for each particular situation. Additional recommendations are for a specialized platform, to share knowledge on RAS, together with a more in depth and distinctive education programme.

1. Introduction

The lack of space for expansion and new sites (due to competition with other uses and interests), limited fresh water availability, and concerns over pollution are considered as key obstacles for further expansion of conventional cage-based and flow-through (FTS) aquaculture systems. Therefore, European countries –mainly existing aquaculture producers – United Kingdom, Ireland, Italy (Eurostat 2010) and Norway (Bellona-AquaWeb 2009;Eurostat 2011) have promoted Recirculating Aquaculture Systems (RAS) as one of the possible solutions and opportunities to further develop aquaculture. This approach is encouraged also in the European Commission strategy documents (COM 2002;2009).

Several countries among the old continent are moving into RAS systems, justifying their change with sustainability reasons.

In Denmark, for example, which is the “fifth largest exporter of fish in the world” (Ministry of Food 2011), the aquaculture industry is “characterized by recycling systems” (Waterland 2011). The governments’ strategy (Operational Programme for the Development of the Danish Fisheries and Aquaculture Sector 2007-2013) is to increase aquaculture production, whilst reducing nutrient discharges (e.g. nitrogen levels) (Ministry of Food 2007). Here, aquaculture is predominated by the rainbow trout (*Onchorhynchus mykiss*) culture. A recent report (Jokumsen and Svendsen 2010) on the technologies used in Denmark, for the culture of this species, showed that RAS are increasingly important. d’Orbcastel et al. (2009c) noted that “more than 10% of trout was produced in RAS”, as they are considered one of the most sustainable methods of fish production. Already, in the early part of the Century, Blancheton (2000) cited that many of the hatcheries within Europe were using RAS systems, while research projects were under development.

Another clear example is the production of Atlantic salmon, the highest value species for European aquaculture (production of nearly one million metric tonnes, Tm, with a production value of around 575 million € [ECF 2011]); this is mainly produced mainly in Norway, Scotland and the Faroe Islands (Bergheim et al. 2009). The tendency for future developments in the northwest Europe is to change current flow-through hatchery systems into RAS; in the Faroe Islands, 100% of that production is carried out by RAS (Bergheim et al. 2009).

Consequently, a clear example of new aquaculture industry development region is located the Basque Country (an autonomous community, located in the north of Spain). Here, the environmental conditions are not suitable for cage farming and a lack of space along the coast is an obstacle. Thus, RAS systems have been presented within the “Strategical Plan for Aquaculture Development 2007-2013”, as the main option to develop the fish-farming industry (EG-GV, 2008). More recently, in 2010, a new RAS facility was opened in Getaria (within the Structural Funds for Fisheries programme [EFF]).

Although, as shown in European countries, the development of RAS is positive (in 1986 just 300 tonne/year were produced in the Netherlands whilst, in 2009, the different countries contributed to the production of more than 23,463 t/year [dates derived from Martins et al. 2010]), many systems had been affected badly by poor management or by poor designs. Both advantages and disadvantages have been published by several authors, over the years (e.g. Liao and Mayo 1974;Sheperd and Bromage 1988; Blancheton 2000;Lekang 2007;Timmons and Ebeling 2010). However, few publications have arisen regarding the issues and constraints the systems experience, with respect to management.

RAS systems were developed as a technology for intensive fish farming, used mainly when water availability is restricted: they enable up to 90-99% of the water to be recycled, through the utilization of many different components. These systems allow the operator greater control over the environmental and water quality parameters, thus enabling optimal conditions for fish culture (Heinen et al. 1996). In contrast, high capital and operational costs as well as the requirement for a very careful management and difficulties in treating the diseases (e.g. Schneider et al. 2006), are the main limitations. Moreover, having water in continuous reuse, constant pumping of new intake water is needed, leading with elevated electricity costs i.e. the higher the water reuse, the more elevated will be the costs (Sheperd and Bromage 1988). Thereafter, RAS systems are not simple systems; they are technology-biology interaction systems, requiring performance monitoring (Lekang 2007). They have benefitted from continuous development (from the simplest path of water treatment until the most sophisticated process) (Muir 1982;Rosenthal 1993); nowadays, they are considered “high-tech” methods.

Within the above framework, most of the research has been directed to improving particular devices, as well as the one best performing individually (e.g. biofilters [Van Rijn 1996; Eding et al. 2006; Summerfelt 2006] and solids removals [Piedrahita et al. 1996;Cripps and Bergheim 2000;Summerfelt and Penne 2005]), to compare different techniques (d’Orbcastel et al. 2009c; Pfeiffer et al. 2011a) and to design entire systems based on particular assumptions (Morey 2009). Such approaches almost always focus upon their environmental impact (latest publication Martins et al. 2010) and on pilot-scale trials. In the same way, little has been done to describe potential risks (e.g. Hrubec et al. 1996) and issues (reported failures are for inadequate biofilters use, power failure, bad alarm connection, poor marketing approach and off-flavour problems in the harvested fish), whilst managing the system, and how all the components can be combined together. Most of the conclusions and studies relate to specific situations. However, there are not identical systems and it is difficult to use one particular example to construct a good performance RAS (Piedrahita et al. (1996) cited this output of a workshop on Aquaculture Effluent Treatment Systems and Costs, held at Stirling Universty [June, 1994]). The understanding of the system is one of the key factors in its management, as this requires interaction between engineering and life organism biology and husbandry. One of the most critical parameters reported in intensive farming has been the oxygen demand and its availability (concentration). While this decreases, other unwanted water quality parameter concentrations increase (Piedrahita et al. 1996); and their balance can be achieved only through correlated work between good designs (engineering) and on understanding of animal behavior (Lekang 2007). The work is more accurate and a profitable work if all parameters are monitorized and followed strictly, during the entire production cycle.

The core objective of the present study is to analyze the most important issues, taking/abstracting information/knowledge and experience from both successful and closed companies, from researchers and aquaculture consultants, as well as from the system designers. This overall view will aid in the understanding of where improvements can be made, that will benefit the entire industry.

2. Methodology

A survey was undertaken in such a way as to obtain both quantitative and qualitative data, seeking to analyze both internal and external opinions and experiences surrounding RAS application within the industry. Within the framework of new technologies gaining more importance, a wide range of communication channels were used to reach different interviewees. The idea was to conclude with an overall point of view of the questions presented, in order to obtain heterogeneous results and discussion. Two sides of the industry were distinguished: RAS system companies and producers; on the other hand researchers, consultants and manufacturers. Therefore, two kinds of questionnaires were developed and used, as appropriate, for each of the interviewees: a RAS questionnaire (Appendix 5.1) and a research questionnaire (Appendix 5.2).

The first was directed towards to reference aquaculture production companies. Its main objective was to investigate the practical and implementation side of the industry. Questions about problems that had affected their system (e.g. types and sources of problems) were asked, how they were solved or managed and how these influenced production and economic performance. Since system components and design were/are selected depending upon the site, cultured species, type of water, and life stage, an appreciation of overall system design and context is essential to link the cause and its subsequent effect. General data such as cultured species, produced life stage, system components and more detailed data such as production or working procedure, systems´ monitoring level, disease issues, detailed problem examples and economic impacts were sought. In the last part, opinions were asked on future expectations and development plans.

The second questionnaire was developed to investigate the opinions and experience of designers, suppliers and other advisers on RAS, who are not managing commercial-scale production systems; thus, compare and contrast diverse ideas and approaches for the future. More subjective than the previous one, respondents were expected to draw on knowledge of a wider range of systems, rather than one specific system. The recipients were asked: which of the component was most difficult to handle for a manager and why; the most common and the worst failures in a RAS system, and their proposed solutions; and, finally, the needed (but lacking) information around this kind of system.

Diverse methods were used to involve as many people as possible, with different opinions, involved in the survey. The RAS questionnaire was launched online via “Bristol University Survey Service” as part of the university´s utilities Companies were approached to participate in the survey, after searching for them via the Internet, e.g. viewing each country´s government´s websites and approaching different experts within the industry. At the same time, a link to the survey was posted in several social networks and websites (e.g. European Aquaculture Society -EAS- membership forum, LinkedIn, Aquaculture hub, University of Stirling – Institute of Aquaculture website front-page). In addition, confidential interviews were undertaken together with production managers from different farms in different countries and to experts with different backgrounds (e.g. consultants, researchers, and system suppliers).

Previously distinguished groups, both producers and experts, were analyzed separately: the "Bristol University survey service" was used to analyze the RAS questionnaire, whilst NVivo 9 software was used to analyze the research questionnaire. The "Bristol University survey service" recorded the results in the system, for subsequent analysis of the data. The service permits making both quantitative (e.g. the percentage of people who responded to each option) and qualitative analyses (e.g. cross-tabulate results between two specific questions, cross-tabulated results between a specific question and the whole survey, or additional analysis like word clouding - up-scale words from a certain question answers depending its important, weighted by the number of times appeared -). The interviews, once recorded, were transcribed and exported to the NVivo 9 program. This served to analyze and identify the main ideas, permitting the classification of data following different criteria (e.g. the role in industry or type of working field), summarizing all the answers for each of the questions and creating "mind maps" for more visual and easy to understand results.

3. Results

Replies from aquaculture production companies were not as expected; although, overall, they represent the highest percentage (Table 5.1). Such numbers make clear a) the excessive confidentiality that surrounds the RAS system industry (regarding to their design and operational methods) and b) the lack of interest supporting the study, as many refusals to cooperate were received. The lack of a specific data compilation of RAS systems companies in Europe (corroborating the statement made by Martins et al. 2010 stated) made it difficult to locate and contact them all.

Table 5.1. Classification of number of respondents to questionnaire

	Contacted	Answer/replies	%of respondents
Production companies	36	16+1(*)	46
Suppliers/consultants (**)	90	18	20
Researchers (***)	50	12	24

Notes:
(*) 16 out of 17 producers are from Europe Thus, to undertake a more objective discussion, the last will not be taken into account, for the quantitative analysis of this project. However, it will be used for qualitative data.
(**) Consultants and suppliers are considered to be in the same area as, in most of cases, suppliers also undertake consultancy work.
(***) For the purpose of this project, researchers are considered as individuals working in a university, in R+D areas in different countries and those who have a background publishing research papers in aquaculture.

In Figure 5.1 are shown the sampled top reference companies differentiated by nationality whilst in Figure 5.2 the distribution is made depending on the specie the companies' culture or produce. The highest number of companies is from the UK, followed by both Spain and France.

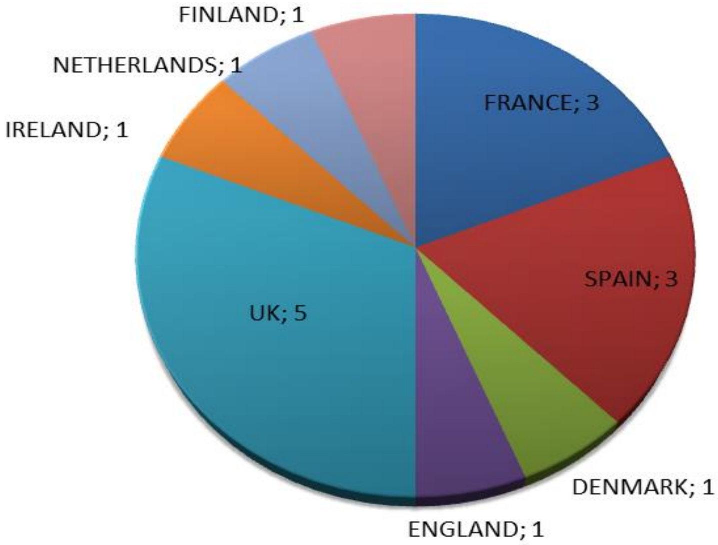

Figure 5.1 Differentiation of the companies participating in the present study, by nationality (showed in %).

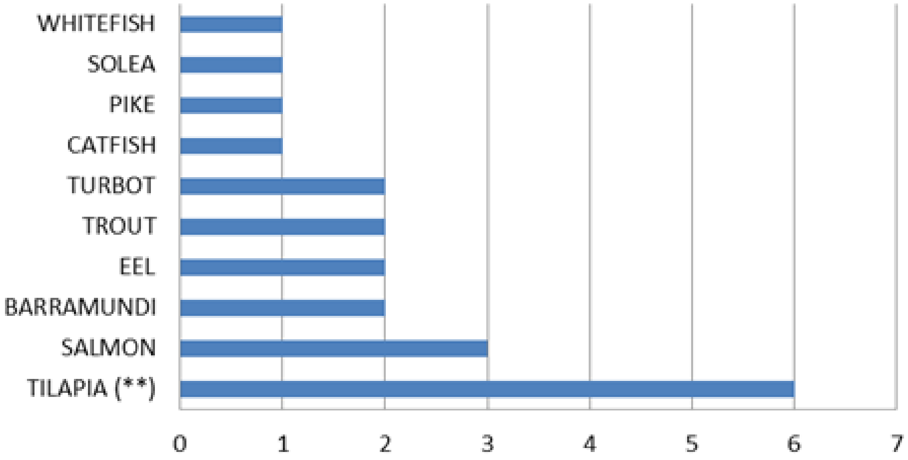

Figure 5.2 Number of companies differentiated by the farmed specie. Note: *The number of companies is not equivalent to the number of species, because some farms are culturing more than one species. **As tilapia are considered as two different genera: *Oreochronis niloticus* and *Oreochronis mossambicus*

These data could assist in updating the research carried out by Martins et al. (2010). The number of companies producing tilapia was the most common (6 companies, representing 37.5%). Thus, 75% of the companies use freshwater (e.g. river or lake water, municipality water, rain water), 18.75% seawater and 6.25% brackish water (depending on species and source of water). Due to the wide variety of species produced, but only limited companies for each, no comparison can be made in terms of management procedures, as well as in terms of failure reasons and financial aspects. Fish life stage is one of the most significant contrasting factors, when classifying and describing different kinds of RAS companies. Thus, in Figure 5.3 respondents are distinguished in terms of the life stage of their culture. From this Figure it can be concluded that most of the production companies that answered the survey are on-growing fish, followed by hatchery farms. Among the 12 on-growing farms, 2 were closed presently whilst one would be reopened in the near future due critic engineering failures. Of the others, the systems of 5 companies were set up as new projects whilst 4 were change to improve the previous systems. The main changes were due to redesigns, from flow-through (FTS) to RAS; also, to aquaponic systems, for different reasons.

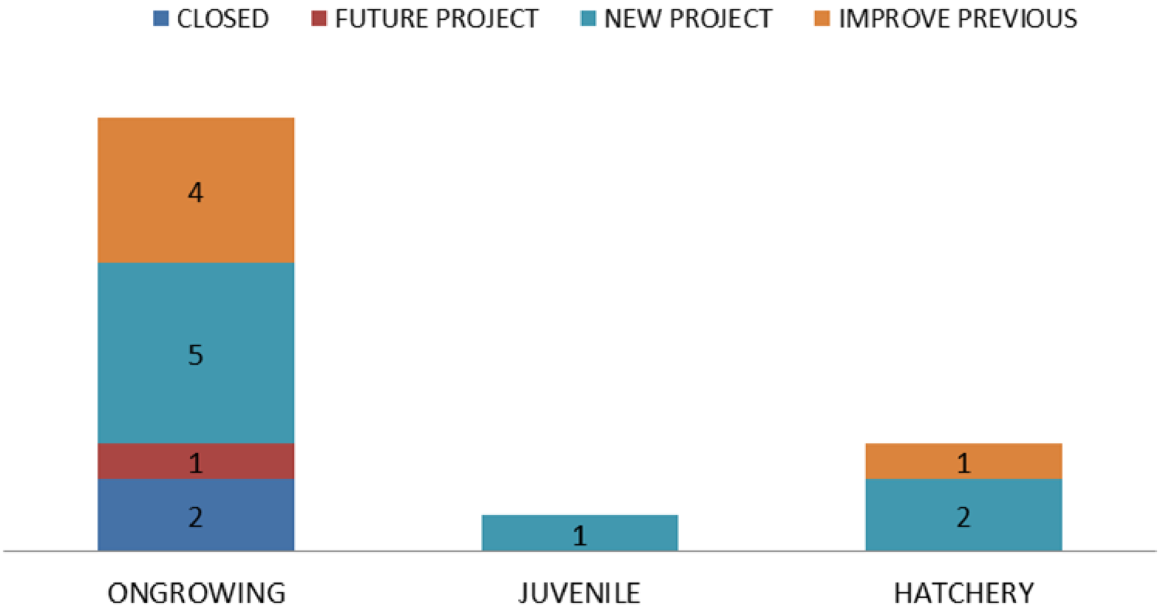

Figure 5.3. RAS systems presented in terms of cultured life stages and current operational status.

Finally, the companies are profiled in terms of the RAS system components used, in Figure 5.4. As can be seen, biofilters and pumps are parts of all systems and solids removal and oxygenators are components for nearly all the systems (94.1% and 88.2%, respectively). It can be seen that skimmers (64.7%) and disinfection devices (ozone is used mainly in all of the seawater companies) are not very usual and neither are denitrification devices (just in 25% of freshwater systems). Within each component category there are different types: e.g. trickling biofilters are the most expanded type of biological filtration devices and drum filters are the most expanded ones for solids removal. For carbon dioxide

(CO_2) removal, ventilators, airlifts and the same biofilters are being extensively used. Heating and cooling methods vary from the use of traditional heaters (gas boilers) and solar panels (photovoltaic panels providing electricity and then used for heating or cooling), to the recovery of energy from the freezers installed in the companies and the use of submerged pumps (also considered a source of heat).

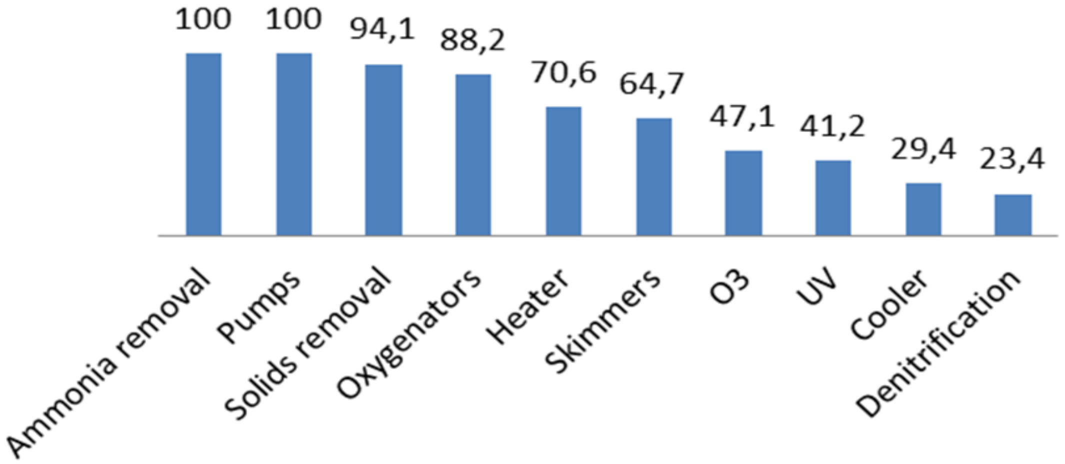

Figure 5.4. RAS components, % of appearance within the companies. Percentage of companies using different water treatment devices.

3.1 Main issues of RAS systems

As cited above, the technology is very dependent upon the life stage of the cultured animal, e.g. it is different to manage newly hatched or small size animals; this is why on-growing and hatchery are considered separately, from here onwards. Cross-tabulating certain questions of the questionnaire it was shown that issues are dissimilar between them. In any case, it is difficult to assess the exact cause of each problem, as the information provided by the producers is not sufficiently detailed and different sources could result in the same consequence (specific examples given are shown in Appendix 5.3). For instance, water quality issues caused mainly by mechanical problems are usual in hatcheries (3 out of 3), whilst badly designed equipment is the most common cause of problems for on-growing systems (5 out of 6). Moreover, whether referring to biological or management problems (i.e. internal or external causes), the answers obtained reveal that issues arise from an initial poor design. For researchers and consultants, clustering the most common issues cited indicates in this order, the main weaknesses: wrong system approach (i.e. inaccurate parameter design calculations, and being too optimistic); inappropriate management (including lack of training); maintenance issues (poor water qualities achieved); and poor system designs (e.g. equipment selection). Likewise, the lack of response to unforeseen circumstances is also a common issue.

Water quality issues´ sources are difficult to assess, as they are produced by different causes: e.g. poor approach of the overall system and production quantities (e.g. lower stocking densities than the real ones used for the calculations); equipment´ failure (in most of the cases due to bad designs); or poor maintenance of the system. Among all the water parameters, ammonia (appearance in 49.06% of the answers), carbon dioxide (25.67%) and oxygen (31.25%) are, for the managers, the most difficult ones to control (results obtained from word frequency query, whilst examining which parameters are monitorized and which of them are the most difficult to control). These are all caused by: (I) a considerable lack of knowledge (followed by complex designs, which is inversely related) and (II) deficient or poor training of the managers; not being able to maintain water quality parameters (with an influence in the performing of both biofilter and solid removal device) (Fig.5.5). Figure 5.5 presents the answers obtained from researchers and consultants (based upon their experiences). Managers of the farms attribute these problems to incorrect specifications in the case of the solids removal device, together with undersized biofilters that rapidly clog. Adding the difficulties of managing certain devices, to the inadequate knowledge and skills of the managers, the final result is an imbalance of water parameters, damaging both cultured fish and the water´s treatment components.

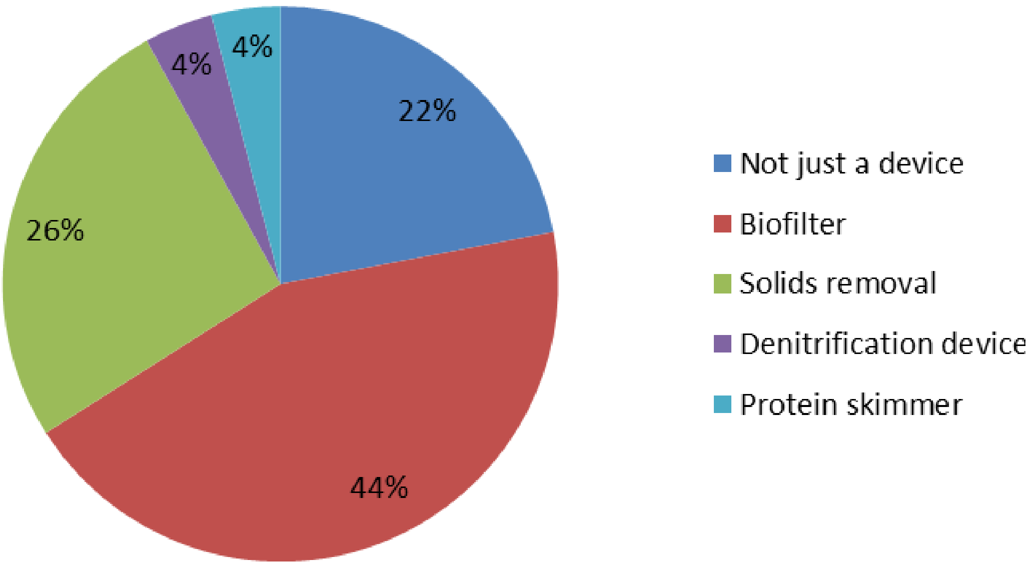

Figure 5.5 Most difficult devices to manage within a RAS system, according to researchers and consultants.

Oxygen and carbon dioxide are also risk factors. Gas imbalance in the system is due to bad designs (e.g. wrong design calculations, inefficient gas stripper, or lack of it) influencing directly carbon dioxide concentrations. Nevertheless, the most common water quality issues (stated by 14/16 companies surveyed and noted by more than two thirds of the researchers and consultants interviewed) were solids in the water, which impact upon the overall system. Most experts consulted agreed that if they are not removed efficiently from the system, the biofilter is affected and does not function properly (i.e. it gets blocked/clogged); thus, nitrification is not completed, leading to high concentrations of toxic compounds (ammonia and nitrite), affecting fish health and welfare.

Likewise, poor initial design, or incorrect assumptions such as assuming lower stocking densities than are actually used, or modeling with simple equations (e.g. kg of oxygen needed per kg of feed), having a substantial impact on final water quality and operational costs (i.e. fish poorer food conversion ratios, increasing solids concentration, ending up with a clogged biofilter). As stated by researchers, RAS systems do not only contain populations of fish, but their effective operation is also contingent upon a thriving population of bacteria: these bacteria consume oxygen and produce waste, whilst their metabolism is vital to the success of the system. This fact is often overlooked by RAS companies; and as such it is one of the worst mistakes leading to failure of a RAS system.

Mechanical problems are also common in hatcheries and on-growing systems, derived, in the first place, from bad design or bad management (i.e. resulting from unexpected conditions). This pattern is created because consultants and suppliers specify that the cheapest equipments are used to meet the demands of the producers for low capital investments. The solutions given for this problems are quick repairs and in last resort replacements. Indeed, this extra capital expenditure due to rapid repairs and replacement were the reason that leaded to some farms to close the business operation. Typically, the most replaced devices, due to a RAS failure, are disinfection devices (i.e. ozone and UV), pumps and biofilters (e.g. 50% of the times when a biofilter or a pump has been replaced, it was for a RAS deficiency, 75% for O_3 and 66% for UV devices). Moreover the connecting pipework and drainage pipes had also been reported as being problematic, undersized and not effectively designed (e.g. slope), respectively. Issues included here directly affect the oxygen amount in the tanks. Another effect is that lower water velocities cause the settlement of solids and/or growth of weed, i.e., compromising the water quality. As an outcome, eleven out of seventeen companies were rebuilt or redesigned completely, following their initial installation; 50% of them due to deficiencies in RAS, whilst the other 50% mainly to extend the production capacity.

With reference to system components, according to few consultants surveyed, biofilters and solids removal are by far the most important, in order to optimize water quality (i.e. for healthy fish and good system performance). However, as the solids concentration increases within the system, increasing fish susceptibility to stress (higher FCRs are obtained, with slower growth) and increasing carbon dioxide concentrations to risky levels, the CO_2 removal becomes a relevant aspect, sometimes not considered, at the designing stage; CO_2 devices are missing in nearly half of the systems, as unforeseen situations and risks are ignored by the designers or installers, when calculations are. An inadequate control over water temperature and the absence of pH control are also identified issues for some systems; among the mentioned causes the inadequate calculations, perhaps based upon laboratory and small scale or trials results are highlighted. One of the most reported issues, particularly affecting on-growing systems (as they produce fish directly for the market) is, off-flavours`. Five out of seven on-growing companies reported that this has been a problem, although the product is depurated, over between two days and six weeks, before sale.

Regarding emergency systems (including both alarm and emergency equipment), two thirds of the consultants agree that poor backup systems still remain in many production companies (the main reason being the desire to have a low initial investment). In terms of emergency equipment, nearly 40% of on-growing producers have just one biofilter and 50% just one solid removal device; this illustrates that little is invested on them. Moreover, in order to decrease the investment, consultants agree that fewer tanks than are really needed (e.g. for the daily procedures such as grading, harvesting and cleaning) and smaller pipe diameters are installed frequently; these compromise daily tasks and increase the probability of failure. Regarding alarms and asking consultants about them, 15 out of 18 agreed that poor alarm networks are in place (in relation to poor or non-maintenance of the installed systems and to a lack of a proper alarm system). Overall, the survey results show that hatcheries have better backup set-ups than on-growing systems due, probably, to the higher added value of the cultured products.

As stated before, unsuitable designs are frequently reported as a common reason of failure. System design relies often upon engineers with a limited comprehension of the science of RAS. Furthermore, the data provided by the managers are calculated optimistically, so designs may not be realistic. The results from Table 5.2 showed that it is notable that there is a similarity between problems caused by equipment, design and RAS system installers/designers. 70% of the systems designed by an external or separate company had problems at some point, whilst none of the farms designed by the final operators reported equipment failures. As reported by the surveyed participants, consultancy support after the implementation of RAS system, from an independent designer, is not as good as is needed (conclusion, 60% of the companies confirm not having an adequate after-sales assistance and support). This is endorsed by the interviewed consultants, who say that many suppliers promise consultancy support availability after selling the product but, in reality, this is limited. Therefore companies need to pay high fees for advice and problem solving.

Table 5.2. Design source of production Ccmpany's system and indications of satisfaction

System designed by:		Separate company	Themselves	With some assistance
Nº of companies		10	5	1
% of the total		58.8	29.4	6.9
Mechanical issues experienced	Yes	7	2	1
	No	3	3	0
Good after sales assistance	Yes	6	-	1
	No	4	-	0

When asking company managers about information available or presently published literature about RAS systems, 9 of them agreed that there is a need for more data and accessible literature; however, they remarked also that this will not be the only solution mostly because, as well as theorical knowledge, experience and practice are needed. 82.4% of the companies agree that there is a necessity for better training, as the current provision is lacking. Moreover, consistent with the views with consultants, all of them admit that it is one of the most important aspects of implementing a RAS. Figure 5.6 shows the areas the information is lacking; hence, where the research should be targeted.

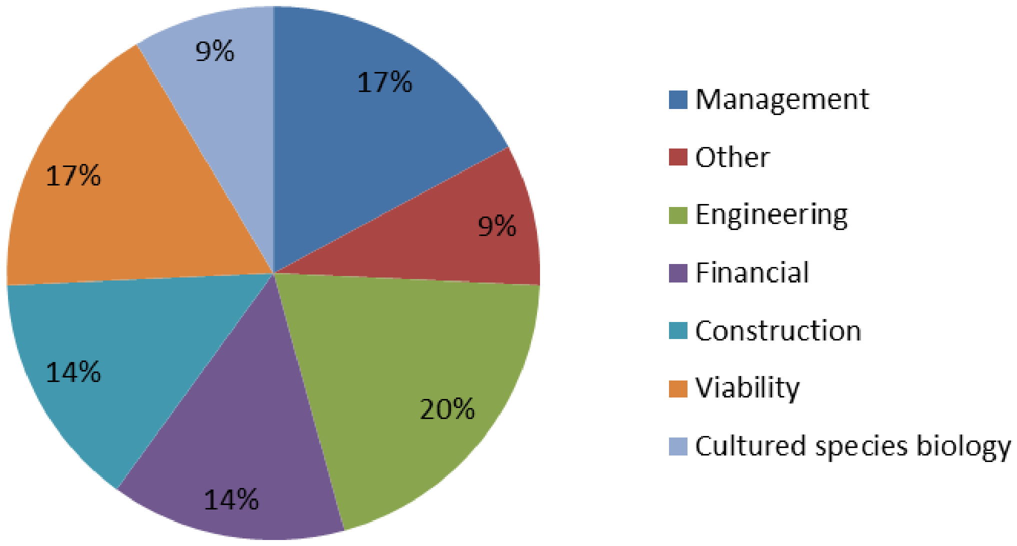

Figure 5.6. Information needs and research areas currently identified as crucial (results from the on-line survey with the production companies, when asked about the lack of technical information on RAS to be developed). Note: numbers appearing in the figure represent the frequency that the particular area has been reported by the companies.

Conversely, looking at the answers of researchers and consultants, there is no need for more information or literature on individual components, what is needed is the improvement of the overall approach to RAS system design (not just technical feasibility, but also economic feasibility) and improvements in design calculations (being more realistic and less idealistic and having in mind that the system can go wrong). More specifically, among the researchers some particular aspects for improvement were mentioned: the understanding of nitrification and, in particular, denitrification, management of produced sludge and the control of off-flavours. Both of the groups agree that there are many people with knowledge in general aquaculture but not in RAS in particular; consultants and researchers blame this on the lack of communication between universities, R&D facilities and companies. It was also agreed that training has to include not just basic water reuse system´s management, but also develop an understanding of the interactions between biology, chemistry, physics, engineering and economics.

3.3 Challenges and future adoption of RAS systems

Finally, financial aspects of RAS were the major issue in response to asking about the challenges to wider adoption in the future. This observation was reinforced by the companies, showing that the financial performance is inadequate in more than 80% of the cases and there is inadequate return on the capital employed, i.e. more than 8 years are needed, on average, to get back the initial investment (Figure 5.7). Therefore, there is a need to reduce costs per unit of production capacity and operating costs. The development of new energy sources and the reuse of system´s byproducts are the main ideas for future development (these appear in 85% of the interviewees answers, as possible solutions).

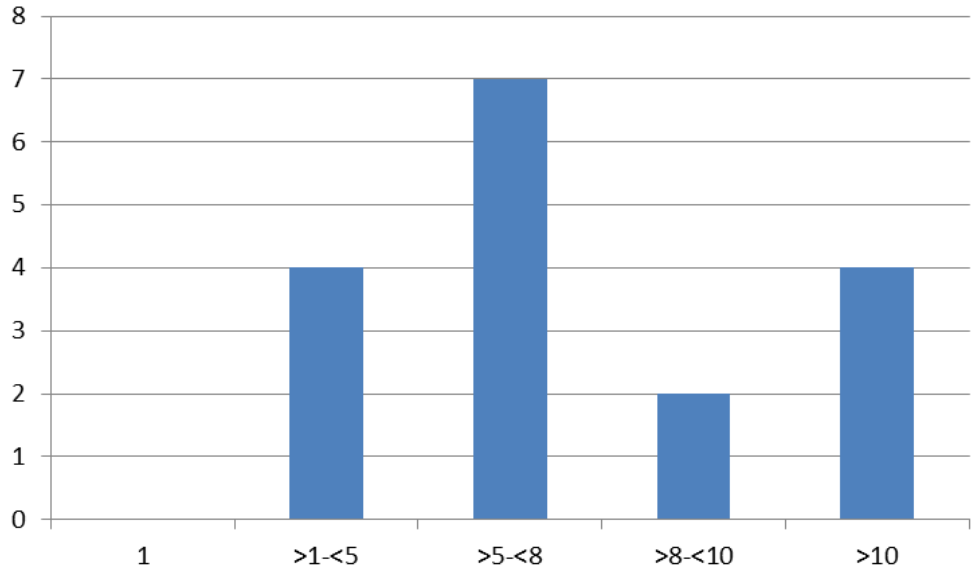

Figure. 5.7. Number of companies per each nº of year frame for the return of the 1st investment.

4. Discussion

The future of aquaculture is to produce fish in a more sustainable way, because demand is likely to increase (FAO 2010) and policy frameworks are becoming more restrictive environmentally. However, RAS technology should secure the control of water quality parameters and the optimization of rearing conditions at the lowest environmental cost. Despite that, the benefits of RAS will depend upon the type and where they are set up. A full control of (I) water quality parameters and (II) water treatment units' performance, to achieve biosecurity levels and reduce environmental impacts, should represent the main benefit of RAS. Nevertheless, their adoption in the future will be determined by the response of industry to the challenges that they face. In the first instance, research and improvements, in terms of individual devices, should be directed towards commercial scale aquaculture, obtaining more reliable and useful data. Their operational systems will need to be better understood, in order to move towards a standardization of the industry. Moreover, in terms of improving their management and having more efficient and less failure prone systems, more specialized and highly capable people will need to be trained. By now, more than 50% of the companies surveyed have been rebuilt or redesigned due to RAS system´s failure. As stated within this contribution, many are the factors and interactions, from the designing stage through the product quality, which can affect both the production success and the subsequent economic profitability of the selected business concept using RAS technologies (Fig.5.8).

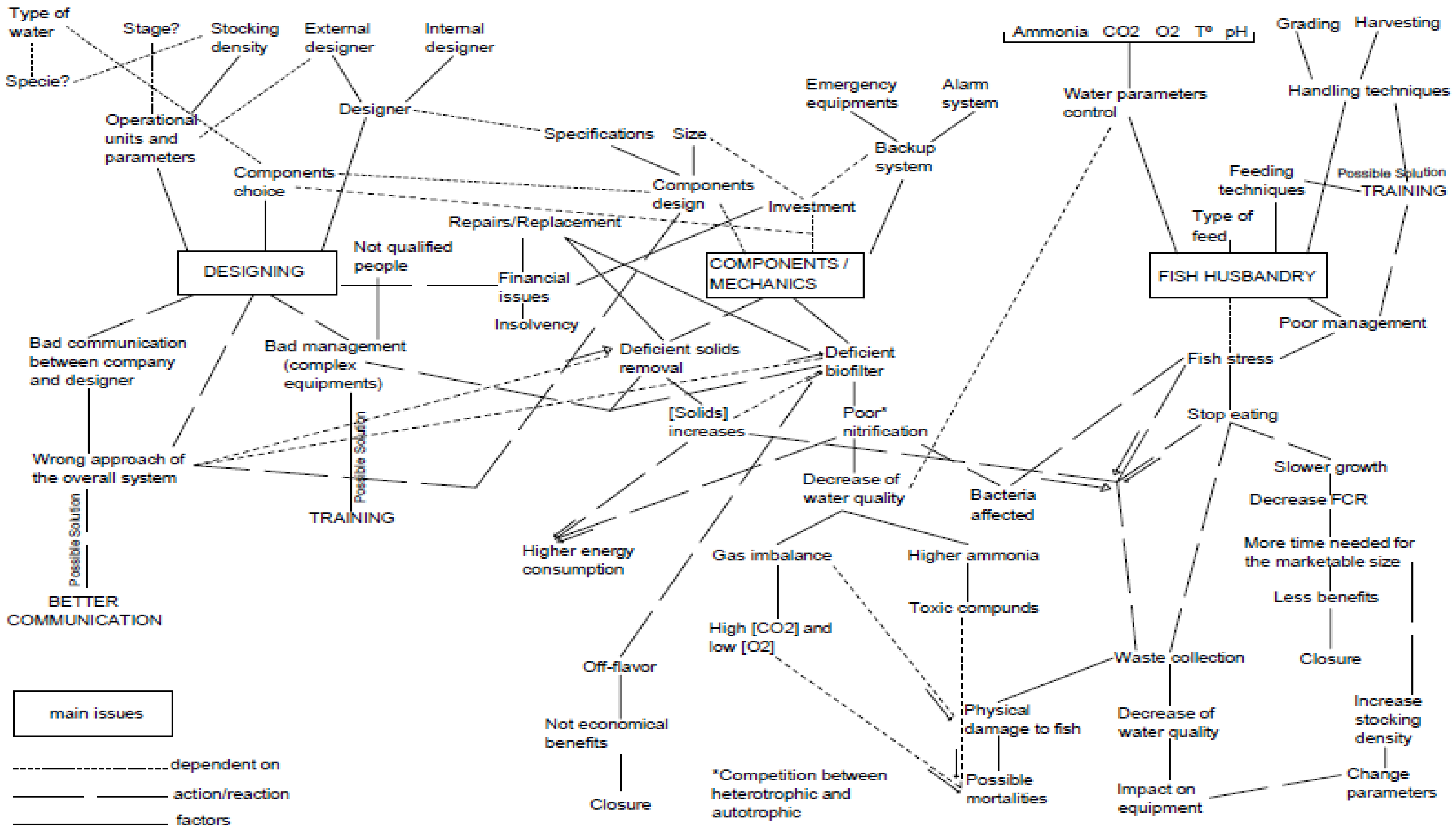

Figure 5.8 Mind map representing factors and interactions, affecting both the production success and the economic profitability of the selected business concept, considering lifespan stages from the RAS designing until the product quality.

4.1 Main issues of RAS systems

As reported, solids management and biofilter operation and management are the most difficult tasks in a RAS, constituting the main reasons for system failures. Treatment technology is developed already but how to integrate it all together in the optimum way is likely missing. Rather than looking for better and more complex designs which can often be more difficult to manage, the necessity is to understand which factors are key in each particular system (e.g. fish requirements, energy requirements, water availability). Accordingly to McKindsey et al. (2006), in order to understand each system´s limits, it is required to define physical, environmental, production and social carrying capacity issues; this argument will ensure consistency in meeting the required sustainability needs of the commercial production systems using RAS.

Suspended solids are the source of most of the water quality issues, as they have an important impact on the performance of nearly all of the other RAS components as shown by the present study; therefore, their management is fundamental for the systems good performance as stated already by Han et al. (1996). A biofilter is affected directly if suspended solids are not removed efficiently from the treatment loop (e.g. Jokumsen and Svendsen 2010); it becomes clogged, decreasing its specific surface area (SSA)[1] and, thus, the quantity and the viability of living bacteria. Moreover, as the solids concentration increases within the system, water parameters are modified and these changes are the causes of stress in both cultured fish and nitrite-oxidizing bacteria (Malone and Pfeiffer 2006; Emparanza 2009), hampering their performance due to their susceptibility to changeable situations (Singh et al. 1999). At the same time, inadequate solids removal creates a competition between both heterotrophic and autotrophic bacteria (Satoh et al. 2000; Zhu and Chen 2001; Leonard et al. 2002; Ling and Chen 2005; Michaud et al. 2006), increasing ammonia levels in the water amongst other things. Apart from the biofilter, other equipment, such as ozone devices and pumps, are also influenced. Ozonation becomes less efficient as the solids concentration increases (e.g. when feeding spikes occur during the cycle) (Summerfelt et al. 2009a) in the water; this necessitates a longer contact time to destroy particulates, which can lead to production of more dangerous O_3 byproducts as the concentration increases. At the same time, suspended solids cause mechanical issues in both of the equipments cited, which can lead to the need for repairs and, thus, additional costs, as reported in the present study. Therefore, suspended solids extraction from the system has to be rapid and with as little breakdown as possible, by not treating them harshly (Summerfelt et al. 2001; McMillan et al. 2003). Further research should be targeted at improving their removal using different kinds and combinations of methods; nevertheless, this will need to be at a commercial scale. However, any combination of the components must be suitable for the farmed fish species and their particular water quality requirements, as well as in accordance with the cost efficiency. A good solids removal management strategy will be necessary also to control the microbial community of the system, thus ensuring a properly functioning biofilter. Accordingly, this has begun to be investigated in recent years by Davidson and Summerfelt (2005); Couturier et al. (2009) and Ray et al. (2010), who showed that a "polishing unit designed specifically to remove fine particles" is needed, in order to capture up to 95% of the solids and, therefore, improve a system´s efficiency; however, in those experiments, the component´s contribution to the whole system´s performance varied, showing different results and requiring further research into the future. However, as reported by different authors, the use of micro screens drum filters seem to be a cost-effective type of solids filters in the classic range of 40 to 90 micron filtration (Carlsen 2008).

Together with solids removal devices, biofilters constitute a non-less important and difficult device for management. A good understanding of both biofiltering operation and maintenance requirements is essential. However, as reported by different authors and also concluded herein one of the reasons for biofilters being difficult to manage is because investigations until now have been focused upon laboratory scale trials, whilst it has been shown that commercial scale RAS waste (more feed inputs, creating higher organic carbon concentrations) is very dissimilar to that produced in pilot scale (Zhu and Chen 1999; Losordo and Hobbs 2000; Ling and Chen 2005; Emparanza 2009; Guerdat et al. 2010; 2011). Thus, as 85% of the interviewees support, more information about the impact of organic compounds on the biofilters is needed in commercial scale systems, as there is only limited data available. Since a biofilter´s characteristics determine the maintenance requirements and management techniques needed the search for standards to classify them and provide specific information to the industry is very likely what the market (companies and consultants) requires. Several authors have addressed already this need (Drennan II et al. 2006; Malone and Pfeiffer 2006; Colt et al. 2006), but once again, little practical on-farm research has been undertaken (Suhr and Pedersen 2010; Guerdat et al. 2010;2011). Apart from this, biofilters rely on many parameters (Chen et al. 2006) and a rapid and accurate actuation is essential, in case of an unexpected imbalance. This approach requires strict working protocols and experienced and knowledgeable management as reported in the present study. There are many complex factors that interact during the commercial operation of a RAS and its biofilter. Daily procedures, such as tank cleaning, grading and harvesting can affect biofilter´s efficiency because water parameters are modified, affecting the hydraulics and causing system fluctuations; similarly, when fish are harvested or removed from the system, for sale, the biomass accordingly declines. Furthermore, the biomass is changing continuously, fish continue to grow whilst more are introduced; this leads to more feed input, higher temperatures (as there is higher metabolic activity), increased carbon dioxide and ammonia production and less oxygen availability (more competition), slowing growth. Therefore management requirements become modified. Thus, managers have to reorganize gradually, to take into account abrupt changes within the biofilter and try to lessen their impacts otherwise both living bacteria and cultured fish will become stressed, leading to uncontrolled system parameters and high fish mortality rates. Some possible management procedures for salmonids, on a commercial scale were presented by Emparanza (2009); it was concluded that feed input, water exchange and stocking density are the variables with the most impact. One reported solution is could be the oversizing of biofilters, to ensure they are more flexible in response to changes; however, this formula demands also higher investments. So that a suitable balance can be reached, calculations need to be more realistic and less optimistic (i.e. including a margin of error) whilst cost-effectiveness needs to be a requisite, in relation to the four types of carrying capacities (physical, production, ecological and social) of the system (McKindsey et al. 2006). Finally, the person in charge should always be able to anticipate required system modifications, understanding relationships and interactions among the parameters, cultured fish and external outputs (i.e. feed, oxygen, energy and water).

As carbon dioxide is produced by fish, its concentration increases where higher stocking densities are used; it causes “uncomfortable situations” in fish, eventually affecting the whole production. However, as stated by companies, equipment for stripping this particular gas (e.g. packed column, agitators) are not used widely in the companies, mainly due to a wrong or poor approach to system design and higher investment requirements. In reality the appearance and subsequent monitoring of abnormal CO_2 concentration could help to more rapidly identify other problems (Pfeiffer et al. 2011a), assisting the better management of the system.

Although off-flavours are not the most common reason of failure in the industry, they can be a motive for bankrupt, because no profits are obtained if fish do not meet consumer demand. It is known that both geosmin and 2-methylisoborneol (MIB) are responsible for this “earthy” and “musty” taste in the products (Tucker 2000; Howgate 2004; Houle et al. 2011) but how to remove them, or how to decrease their occurrence, is still under investigation (Schrader et al. 2010) without much success. Guttman and Vanrijn (2008) have proved that having anaerobic conditions within the system could be a possible solution for the mitigation of this problem. Likewise, denitrification devices, although presently not very common, are being used where high levels of nitrate, high stocking densities and high levels of C/N interact (van Rijn et al. 2006). Thus, adding a non-aerobic denitrification stage after the aerobic nitrification (i.e. biofilter) could likely mitigate both water quality and off-flavours issues at the same time; however, this will need further investigation.

4.2 Challenges and future adoption of RAS systems

One of the greatest reported constraints of RAS is the investment required and the long pay-back periods (on average 8 years). RAS are frequently not economically viable; “encouraging technology” is inevitable, but there must be an economic reason, in relation to an overall “market-need” oriented perspective of the system that ensures technically feasibility as a prerequisite to be economically viable. A good market or social study is needed, in order to meet with the actual demand, planning an affordable and realistic production goal. Thus, the first requirement is a reliable operation followed by low operating costs. Both conditions will aid recover more rapidly from the first investment: the first obtaining a stable production and, thus, profits; and the second providing a higher margin for the return. Some possible ways or solutions, as given by some of the interviewees, to make these systems “cheaper” are listed below; however, they will need to be investigated further, in terms of operational management and economical viability:

- Energy efficiency, using less and reusing energy where possible. Reducing pumping head and improving the biofilter´s performance for instance, means less energy will be needed (Jokumsen and Svendsen 2010).

- Recovering wash water from the drum filter backwashing e.g. using flocculants (currently under investigation) will reduce the amount of intake water, decreasing environmental impact and reducing pumping costs.

- Introduction of new compartments such as algal and for aquaponics production to (I) decrease environmental output, (II) valorize nutrients and detritivores taking advantage of produced byproducts such as carbon dioxide and (III) generate secondly products to a major economical input.

- The implementation of a “hybrid technology of biofloc technology (BFT) and RAS” as Azim and Little (2008) suggested. A more recent study showed that BFT could help environmental and economical sustainability of RAS by reducing the feed cost (Kuhn et al. 2009).

It is generally accepted that Europe has the advantage of having the technology and the knowledge needed to set up RAS (COM 2009), but this technology is more than just turning an “on/off” button and leaving it to run; it takes time to learn how to manage it. The systems are complex, in terms of understanding how they need to be handled in each particular operation situation; they depend upon many parameters which, in turn, depend upon the performance of each of the constituent parts. As stated by the interviewed participants, people with the responsibility of managing recirculation systems should be trained with functional skills, within university educational programs and on further practice or internships within research and/or participative production companies.

Fish farming is necessary and more will be needed in the future. Hence, RAS systems will continue to develop, but their improvement cannot be achieved if there is no communication within the industry (involving producers, suppliers, researchers and consultants). Furthermore, it is well known that the lack of information is due to a lack of governance (e.g. APROMAR 2010; Scottish 2003), together with and insufficient collaboration within different work areas in aquaculture. Thus, as concluded for this study there is a disincentive for communication at a commercial level, as well as a fear of reporting “bad news of failures” to the public. Nonetheless, knowledge of RAS control and management techniques are gained with experience and, as has been demonstrated, a knowledge of the technical or engineering part of the system does not always lead to success. Moreover, this study has shown that suppliers and producers do not agree, when requesting industry´s point of view, revealing evidence of individualism. It is considered (and confirmed herein) that sharing experiences and issues (without compromising on confidential data), can be beneficial for all parties. This study has confirmed also that social networks are useful communication channels and they are nowadays the best way to bring the people studying on RAS together.

Appendix 5.1 RAS Questionnaire

The core goal is to analyze the most important issues for the management of recirculated aquaculture systems (RAS). We hope that learning about problems in the past from many different kinds of systems, will help to identify priorities for future research and training that will lead to better solutions for future managers. This information will aid understanding of where improvements can be made that benefit the entire industry. All information provided will be treated as confidential and individual operations will not be identified in any reporting.

Company´s name:
Name of Producer:

Contact details – Address:

Telephone:
E-mail:

Species produced:

General Information

- Species cultured and life stages:
- Seawater ☐ freshwater ☐ Brackish water ☐
- The setting up of the RAS system was (tick the best option):

  New project ☐ Improve previous ☐

  If second option, where did you make the improvements?

- Has the farm been extended after the initial installation? Yes ☐ No ☐

If yes, has it been modified or upgraded due to deficiencies in the initial design?

Yes ☐ No ☐

- How monitorized is the system? (i.e. just the tanks monitorized or you also have video cameras, flow meter, water quality parameters monitorizing, light level sensor, system biomass sensor)

  Entirely ☐ Partially ☐

Production

- Production of the year 2011 (Kg/year):
- Was this production the optimum of the system?
- Continuous production ☐ batch production ☐
- Have you ever had mortalities due to a failure of the RAS system?

  Yes ☐ No ☐

- Do you regularly suffer higher than expected mortalities due to using RAS system?

  Yes ☐ No ☐

Water system

- What the distance to the intake water source? And how is it taken?
- Do you need to keep topping up the system´s water level? Yes ☐ No ☐

If yes, what percentage of the total volume of the system do you top up?

- When you add new water to the system, does it enter directly or does it go through the treatment phase first?
- Do you heat up or insolate the building to keep the temperature of the production unit? Yes ☐ No ☐
- Do you hold a reservoir or store water which is available in case of emergency?

Yes ☐ No ☐

If yes, have you ever had to use it? Yes ☐ No ☐

- What water quality parameters do you measure? And how often?

Infrastructure – system components

- Do you have emergency power generator? Yes ☐ No ☐
- When were the (main) components of the system installed?

Beginning ☐ following time ☐

If the "following time" option, why? Just improvement ☐ Something wrong ☐

- Have any components been replaced? Yes ☐ No ☐

If Yes what components?

- Have you ever had to take the biofilter out of service during a production cycle?

Yes ☐ No ☐

- And any other component?
- How often is the biofilter cleaned:

at the end of cycles ☐ specific schedule ☐

- Do you think that the biofilter is the main component of a RAS ? Yes ☐ No ☐

If No, which is it and why?

Main discussion

- Was the RAS system designed and installed by

separate company ☐ yourself ☐ with the assistance of any company ☐

If it was for a separate company, do they offer you good advice or help?

- Please rank your top five of the main problems/ issues /challenges in system management (regarding in particular to the RAS issues)

- Main risks faced by the manager?

- Problems that have happened in the past – give some examples and how they were solved
- Average number of RAS problems per annum

- If you have had problems with the RAS system, has it produced mortalities?

Yes ☐ No ☐

- Are RAS problems relatively higher in percentage than any other kind of problems (human errors, administrative, legal, food/safety problems?

Yes ☐ No ☐

System general management

- Have you ever had off-flavour problems? Yes ☐ No ☐
  If yes, in which stage?
  If yes, do you depurate (put in a separated and clean water system) the product before?

- Do you have biosecurity concern? Yes ☐ No ☐
  If yes, what kind of procedure do you have (e.g. no visits allowed, foot disinfection)

- Have you ever had disease problems? Yes ☐ No ☐

- Source of fingerlings: buy from a external source ☐ produce them ☐

  If the first option,

  Do you have any quarantine facility to hold the received animals?

  Yes ☐ No ☐

  If yes, what quarantine facilities and procedures are in place?

- Tick if you have you lost fish due to: Which?

- Mechanical problems	☐	
- Electrical problems	☐	
- Biological problems	☐	
- Operators/human errors	☐	

- How were each of the problems solved?

  Mechanical problems

  Electrical problems

  Biological problems

  Human errors

Other questions and comments

- If you were starting or investing in another project, what modifications, compared with the current system, would you make?

- Is the any lack of information about RAS systems that you think that is crucial?

- Would you like to see more training available on RAS systems? If so, what sort of training and who should it be aimed at?

- Is the financial performance of the system adequate? Do you get a reasonable return on the investment?

- Which do you think that are the key factors for the future development of the RAS systems?

Any other comments or personal experiences

Appendix 5.2 Research Questionnaire

The core goal of the project is to analyze the most important issues for the management of recirculated aquaculture systems (RAS). We hope that learning about problems in the past from many different kinds of systems, will lead to better solutions for future managers. This information will aid understanding of where improvements can be made that benefit the entire industry. All information provided will be treated as confidential and individual operations will not be identified in any reporting.

1- In your opinion, what is the most important benefit of RAS systems? (i.e that is already driving adoption, or likely to do so in the future)

2- Which component in a RAS system gives the most difficulty to managers, and why?

3- What are the worst failures in RAS systems that you have seen or have been faced with in your experience (e.g. biological, mechanical, human errors etc)? Give particular examples if possible.

4- And which are the most common failures you have seen or been faced with in a RAS system? (make a kind of ranking: top ten or top five problems, for example)

5- Having in mind your answers to the previous two questions, what solutions could you give for those problems?

6- The problems you have cited, are caused for a bad management? or because people buy systems without knowing exactly what they are buying?

7- In your opinion, is there a need of more training and more specialized people? How could this be solved? Which kind of training and who aimed at?

8- Which are the most common questions that people (fish farmers, people who buy a system) make about RAS?

9- Is there any lack of information about RAS systems that you think is crucial? E.g. Key things in the system that should be a research target in the future.

10- What developments or changes would enable RAS to be adopted more widely in the future?

11- If you were investing in a new project, would you definitely invest in RAS? Would the answer be the same for any kind of species or life stage?

12- Do you work together with any research company/foundation or in your own?

Other comments or experiences:
*Please, if possible provide a RAS system schematic that you have worked with/on.

Appendix 5.3 Examples of particular RAS issues

The following tables show some particular examples of problems or issues reported by the companies that completed the survey. The problems are differentiated between hatchery and ongrowing systems. At the same time, issues are classified by type.

Hatchery systems

WATER QUALITY ISSUES

CAUSE	EFFECT	SOLUTION	CHANGES IN THE SYSTEM
Zeolite from the sand filter was removing calcium from an already soft water	High rate of mortalities while transferring from the incubation to the first feeding	Remove the zeolite from the sand filter	Increase the biofilter capacity to improve the ammonia removal at low temperatures
Waste collection in the tanks, decreasing water quality	Fungal infection, gut and gill fungus	Treatment	Change temperature regime at 1st feeding, to encourage fish to swim up off from the bottom of the tank
Too high ammonia level in the fish tanks	High rate of mortalities	New water introduced into the system	

MECHANICAL ISSUES

Breakdown of the sand filter: lateral side of the filter came off	Release sand and dirty components into the biofilter	Repair	Lateral system of the sand filter rebuilt, more robust system
Problems with solids removal (pressurized downward flow filter), pumps, oxygenators.			

DESIGN ISSUES

Inadequate CO_2 stripping (wrong or poor operational calculations)	High CO_2 levels in the water, fish getting stressed (less feed intake → more solids in the water)	Repair the gas stripper	Additional CO_2 stripping installed in the biofilter to blow air upwards in the trickling tower
Heat exchanger overheating the biofilter	Biofilter bacteria was being killed	Change the layout of the farm	Redesign the system
Airlift for transporting the water poorly designed	Supersaturation in the tanks (>110 N2 saturation) and fry mortality		

<u>Ongrowing systems</u>

WATER QUALITY ISSUES

CAUSE	EFFECT	SOLUTION	CHANGES IN THE SYSTEM
	Lack of oxygen combined with high nitrogen compounds and CO_2		
Temperature of water drops in winter	Too low T	Increase heating and recover heat from waste through heat exchangers	Improve protocols and system design
High ammonia levels	Biofilter was not working efficiently	Fine tuned filtration system and improved the biofiltration + addition of bicarbonate	Improve protocols and filtration system
High turbidity in the water due to the solids removal device	Biofilter was not working efficiently, it was clogging	Improve drum filter design, getting an smaller mesh size (down to 40 microns)	Improve filtration system
Poor water quality conditions (mainly due to the drum filter)	Low feed intake, decreasing the FCR and producing more waste	Better feed and feeding techniques	Improved type of feed and education of employees on best feeding practices
Poor water quality produced increasing bacteria population	Off-flavour issues	Improving the filtration and purging the fish	improving purging systems

HUMAN ERRORS

Not following the right protocols, unfamiliarity inexperience, i.e. leave a tank without oxygen	Fish start to be lethargic, not eating. More waste was produced	New protocols	New protocols implemented in the farm and a backup system installed
Poor handling	Stress of the animals remaining in the tanks	New protocols and education	
Inexperienced employees, lacking knowledge of correct feeding techniques	Mortalities (after some lesions, fatty livers, bloating)	Change of diet	New protocols and feeding techniques

DESIGN ERRORS

CAUSE	EFFECT	SOLUTION	CHANGES IN THE SYSTEM
The design was not the best	Oxygen was not properly reaching properly the tanks	Pipes and channels were modified, as well as their entrance	
Tanks walls were not high enough	Fish jumping out of the tanks	Increase the wall height of the tanks	
Poor aeration and diffusion	Low oxygen levels in the tanks	Improve equipment/repair	Protocols and improve the system
There was no denitrification devices installed in the system (high densities)	Nitrate levels increased in the system	Denitrification system was implemented	Monitoring nitrate levels both in the inlet and in the outlet of the biofilter
The stocking density was higher than the one used for the calculations	Biofilter could not cope with so much ammonia, it was to fixed design	Installation of a large fluidized bed biofilter	
The stocking density was higher than the one used for the calculations	More oxygen was needed in the system, as well as more ozone	Increase oxygen and ozone production	Installation of a large O_2 and O_3 injection systems, to have plenty
Poor designing of the tanks	Leaks	Re-weld the tanks	Design new tanks
Pipes and channels were not effectively designed, the slope was not enough to move the water at a certain velocity	Settlements/ weed growth occurred in the slow moving waters	Increased the slopes on troughs and channels to facilitate cleaning	
The pipes were not placed adequately.	Too much power consumption	Removed return pipes over the side of the tanks and replaced with return troughs (water entered just 5 cm above water level)	

First design made without external oxygenators and the amount of oxygen needed was higher than expected	Cannot reach the expected production → do not sell any product and do not make money	Close the farm	
Pipe diameter too small	Water did not run as quickly as it should	Quick repairs in first instance	New protocols for maintenance
Few tanks installed in the system (to save money at first investment)	No space for the grading or treatment of the fishes	Spend more money than expected without selling fish	
No cooling system installed	Water temperature increases and the cold water species cultured in the system began to die	Installation of a cooling system (high energy costs)	
Solids removal device undersized	Never achieved the desired density		

MECHANICAL ERRORS

CAUSE	EFFECT	SOLUTION	CHANGES IN THE SYSTEM
Pump failure	Lack of oxygen within the tanks	Change the pump	Install an emergency pump
Cooling apparatus failure during the summer	High temperatures resulted within the system and fish die from a bacterial infection	Repair	
Biofilter crashed	Poor water quality achieved	Change filter media	Backup biofilter installed for the emergencies
UV lamps failure, air pump failure			

General discussion

Various aquaculture investors have shown their interest in commercially producing aquatic species in the Basque region (EJ-GV 2008). Thus, among different and most commercialized fish sold in the local markets, Atlantic cod (*Gadus morhua*) and Atlantic salmon (*Salmo salar*) were the ones selected for the study. The main reasons for such decision were: both species flesh's healthy properties, exponentially increasing consumption trend for salmon during the past years and, the advantages of culturally consumption habit of the region for cod.

Atlantic cod is likely the most popular cold-water marine fish species in the world (Kurlansky 1997). In aquaculture, this species farming potential began when wild individuals were over-exploited (Esmark and Jensen 2004; FAO 2014) although very few are the attempts of on-growing cod in land-based RAS (Lambert and Dutil 2001; Rosenlund et al. 2004; Fülberth et al. 2009). Atlantic salmon's industry for its part, has substantially grown coinciding with the decline of wild stocks (Asche et al. 2013; NASCO 2014; FAO 2016), the rise of both supermarkets' and consumers' interest for a healthy eating life style and, salmon products' attributes and format offers (Forster 2010; Asche and Bjørndal 2011; Seafish 2011). Thus, currently, salmon is the highest-valued species grown in Europe.

Technical feasibility

A RAS farm can be isolated from the surrounding environment; there is no need to set up a farm where favorable conditions exist. Nevertheless, this would increase both economic costs and environmental impacts, requiring a conscientiously high biosecurity levels. Thus, in normal situations, a minimum interaction happens, and it is then when the viability of the system should be proven, and question such as the following arise "*Is the surrounding environment favorable for the producing species?*", "*How can be the RAS adapted in order to take advantage of the conditions around?*", "*Which are the most adequate RAS management strategies?*", "*Would management have any impact in the technical feasibility?*"

Setting up a RAS in the region seems to be technologically feasible, according to the thesis results. It has been proven that an early temperature manipulation towards colder water can lead to a long-term positive growth effect in cod (Luczkovich and Stellwag 1993; Imsland et al. 2005b) and halibut (Larsen et al. 2010), stimulating benefits in commercial aquaculture (e.g. faster growth in a given timeframe). Thus, in the present thesis, a comparison (i.e. growth performance and flesh quality) was made in both cod and salmon experiments between individuals reared at a stepwise (i.e. mimicking natural oceanic conditions and fluctuations) and constant temperature regimes. In both experiments, mortality rates were similar in both regimes although some differences were found in growth performance. Individuals reared at the stepwise regime experienced a halt in growth during warmer water temperatures period arresting their growth, agreeing with the research published by Jobling (1994). However, these individuals were able to adapt to feast-and-famine conditions by showing marked growth spurts, when environmental conditions and food supplies were increased after a period of starvation (Jobling 1994). Therefore, although it has been proven that the rearing water temperature maintenance played an important role in the study, it cannot be assured, but only suggested, that this growth pattern could be due to such manipulation.

Additionally, water temperature maintenance was made with the use of a heat pump and the adoption of different working strategies depending on the season (i.e. partial reuse or RAS). The first one resulted in being the most energy consuming and environmentally unfriendly process of the water treatment loop, while the second one achieved a gradual, but successful, bio-coupling of individuals to the conditions of the new environment being favorable for both systems' functioning and individual's growth performance. Thus, in order to set up a RAS farm in the region, and having in mind the compensatory growth occurred after warmer periods, a balance should be found between both approaches: a possible strategy to rear cold water species would be adopting a partial water reuse modulation strategy during the coldest seawater temperature periods, and a RAS strategy in summer. In this way, heat pump's temperature maintenance could be decreased, reducing both economic costs and environmental impacts.

Regarding other production parameters such as water quality (i.e. dissolved oxygen, ammonia, nitrite, nitrate, pH), they stayed within the limits along cod and salmon experiments. Nevertheless, special attention should be taken into account regarding gas saturation levels when working in RAS in general and cod in particular. High levels of gas caused bubbled eyes in some of the marketable size individuals, which would lead to the decrease of the benefits. In such manner, for the salmon experiment, system's design was modified: (I) some barriers were set up in the expansion tanks decreasing so the impact of the water when entering here, and (II) the distance between the tanks and the expansion tank was increased lowering water's velocity when entering the expansion tank.

From RAS management perspective, they are mechanically sophisticated and biologically complex. Thus, RAS managers should know about the required water treatment processes, the components involved in each process and, the technology behind each component; this requires education, expertise and dedication (Dunning et al. 1998). In fact, the understanding of the system is one of the key factors in its management, as many commercial RAS operations have failed because of component failure due to poor system design and inferior management (Masser et al. 1999; Timmons and Ebeling 2010). As resulted in the worldwide survey made in Chapter 5, solids and biofilter's operation and management are the most difficult tasks, constituting the main reasons for system's failures. Suspended solids were reported as the source of most of the water quality issues; they directly affect both the biofilter and water quality parameters causing stress in cultured fish and nitrite-oxidizing bacteria (Malone and Pfeiffer 2006; Emparanza 2009). Moreover, the mechanical issues generated by the solids in the cited equipment could lead to the need for repairs adding costs to the production. Consequently, universities, post-graduate studies and research technology institutions should work in parallel in order to create RAS expertise; well formed people comprehending engineering, biology, animal husbandry, and water quality and chemistry knowledge.

Environmental feasibility

RAS are the most environmentally friendly fish farming technology that exist nowadays; they have been scored as a “green technology” by the SeaFood Watch program to rear any species in any part of the world (Badiola et al. 2014). As such, from this perspective, setting up a RAS in the region seems to be feasible. Nevertheless, the present thesis has concluded that energy consumption would be an issue. This agrees with the worldwide report made by the author of the present thesis who enforced the idea of requiring the energy use in RAS as an additional environmental evaluation criterion of the technology (Badiola et al. 2014). Moreover, although the energy use is not important within the RAS industry (Contribution 3), it is one of the main constraint for technology’s wider implementation (Colt et al. 2008; Saidu et al. 2012; Liu et al. 2016). Additionally, a little improvement (i.e. use of renewable energy sources) or decrease in such consumption will enhance the overall sustainability of the technology (Badiola et al. 2014). Basque region’s conditions such as the main energy source (i.e. non-renewable energies) and energy’s cost (i.e. €/kWh) make the production costly from both environmental impacts created and operational costs generated. Thus, how could the energy consumption be decreased? Is there any methodology to precisely monitor the energy that could aid taking accurate energy saving measures? What has been studied around the energy use in RAS? Has been any improvement over the years?

Going deeper on this, Contribution 3 resulted in an extensive review of the published data and information about the energy use and different RAS designs efficiencies. Every process requiring energy (i.e. pumping, filtering, biofiltration, and oxygenation) was studied individually, identifying the improvements made over the years. Moreover, available renewable and non-renewable energy sources and their potential usage were reviewed. It was concluded that RAS energy consumption varies between each case study, depending on several factors such as farm’s location, reared species and produced volumes. At the same time, the design of the systems as well as the selection of proper devices is crucial for a successful and efficient production (Contribution 3 and 5). In this manner, comparisons between different RAS should not be generalized in terms of energy consumption but being aware of the kWh/kg consumed would aid to know which percentage of the costs result from energy and where the modifications in the system should be made to decrease such consumption.

Contribution 4 proposes a combination of Life Cycle Assessment (LCA) with energy audits to improve environmental performance of RAS, identify energy consumption and thus, its environmental and monetary effects in order to seek cost reduction. In the studied pilot-scale RAS, heat pump resulted to be the main energy consumer device from the water treatment loop and its consumption showed a temporal variability as result of seawater temperature’s fluctuation. This agrees with the idea that energy consumption in RAS is not constant along the production cycle and it follows a time-based pattern (Ioakeimidis et al. 2013). In this context, the methodology presented seems promising to quantify the existing energy flows in the system. This method provides real data (i.e. system-specific data) aiding to define time-based energy-saving measures from both economic and environmental terms (e.g. CO_2 eq). Moreover, such methodology combination improves the identification of the environmental impacts created by RAS by performing a more complete and precise LCA and eco-designing the fish rearing process. In this particular study, the impact categories assessed in the LCA showed that fossil fuel based on-farm electricity for the on-growing of fish was the most environmentally unfriendly input. This reinforces the idea that an improvement in the energy use (e.g. decreasing the energy needed, using renewable energy sources; optimizing systems) and thus, its consequences (i.e. environmental impacts created and economic expenditures), would be an important way forward towards a comprehensive sustainable technology.

Regarding the energy use in RAS, the location of the farm is an important parameter which may change the created environmental impacts and will be a key parameter in the production's operational costs. On top of this, depending on the country, renewable energy is often sold to clients that pay extra for a certificate to claim that their electricity is produced from renewable sources. The location could make renewable energy sources option to be cost or non-cost effective.

What if I am a RAS manager? The kWh/kg fish produced will aid to know which percentage of the operational costs is due to the energy consumption. Moreover, making an energy audit will let you know which the main energy consumers are and when the consumption peaks occur. This could assist to make a plan and looking for the best available energy source option in each moment. Currently, non-renewable energy sources, which increase CO_2 emissions in comparison to renewable sources, are the most preferred ones within the industry. In contrast, renewable energies clearly may decrease the greenhouse gas emissions being an inherent alternative to consider. Nevertheless, in general, RAS companies' electricity is generated in a public utility (i.e. dictating the energy costs, $ or € /kWh), limiting the options of the energy source. Moreover, nowadays in Spain, the electric power is a commodity in short supply and power markets are connected through economy and/or the grid, making challenging the inclusion of alternative energy sources. In this manner, the unique choice for the use of renewable energy sources would come if RAS managers decides to generate the electricity independent from an utility company (i.e. when a public utility is unavailable or unreliable). This option would be included in the produced fish labelling which could be a reason to increase the price, targeting environmentally aware consumers.

What if I am a fish consumer? Seeking information and making questions about the fish you are purchasing in the market would be the first step. As much data the consumer obtains more confident they will be, creating higher demands. At the same time, this interest will slowly but steadily make producers to go towards more friendly productions standardizing "greener productions" (i.e. using more renewable energies) and thus making pressure on their costs (i.e. energy costs). In parallel, the technology used for fish production should be included within the fish labelling and in case of RAS whether the used energy source is renewable or not should be indicated.

Economic feasibility

Obtaining benefits is the main goal of any industry; a balance between what is spent and what is gained. Nevertheless, in RAS, financial aspects are the major issue to their widely future adoption (Contribution 5). For instance, although the number of RAS production companies has increased in the last years (Bergheim et al. 2009; Murray et al. 2014), their high energy demand (i.e. operational costs) is still one of the main disadvantages both environmentally and economically. In more than 80% of the cases studied in the worldwide survey, the financial performance was deficient and there was inadequate return on the capital employed, needing more than 8 years on average to get back to the initial investment. In the present thesis, the economic study undertaken showed high degree of uncertainty regarding the realistic scenarios in the region. Minimum marketable product’s price required to have a viable production resulted to be too high for the current market. The operational costs responsible of that were employees’ salary and production’s energy costs.

Conventional financial and economic analyses have demonstrated a broadly positive impact for many forms of aquaculture, including the more intensive resources demanding systems such as RAS. Thus, the use of economic tools embracing wider measurements of social and environmental costs and benefits might provide different and possibly more critical perspectives. However, although these techniques hold promise for such analyses, their development and application in sectors such as aquaculture are as yet limited. As such, tools and applications of environmental economics should be promoted and ways in which these may be more effectively applied in strategic and local decision making for aquaculture development should be proposed.

Societal feasibility

For any aquaculture technology to be economically viable, it is necessary to integrate the aquatic resources or technologies with the appropriate marketing strategies. Moreover, social acceptability of the produced fish and their preferences with other species or seafood products will aid to predict market’s behaviors. A RAS business needs to produce marketable products accepted by the consumers, in order to be profitable. In fact, inadequate market demand approach could result in a failure production as shown in Contribution 5. Due to the fisheries popularity in the Basque region, aquaculture products are not widely spread. Nevertheless, their presence in the local markets continues increasing. This study has remarked the positive acceptance that consumers would show in relation to a possible marketable locally grown product. Both cod (Contribution 1) and salmon (Contribution 2) resulted to consumers’ liking. The organoleptic tasting carried out revealed that the quality of fish species was equal or of similar characteristics to the samples obtained from the fish markets. Additionally, seafood experts were not able to discriminate between samples or origins (i.e. aquaculture and wild). Similarly, purchasing intention study of salmon resulted to be more likely towards the locally grown fish than the commercial.

In other food categories marketed in the region (i.e. vegetables, dairy products and/or meat), market price seems not to be a limiting factor when it comes to locally grown products. At this point, marketing strategies must take advantage of the specific benefits afforded to the product by RAS including availability of the fish as an absolutely fresh product (alive or fresh dead), certified as unpolluted, and available continuously throughout the year (Van Gorder 1991). Moreover, information strategies such as the SeaFood Watch program of the Monterey Bay Aquarium, could help to provide incentives to consumers to adopt sustainable choices by giving the accessibility to be aware of the whole fish production (e.g. delivering informative flyers) and/or informing about the benefits of purchasing RAS farmed fish (i.e. environmentally friendly). Taking advantage of the benefits offered by RAS could help to introduce such products to different niche markets where there is a willingness to pay a higher price for a higher-standard products.

The whole feasibility study

Overall, the tiered approach proposed in this thesis may be useful to set up a RAS company in the region. It comprehends technical, economic, environmental and social aspects of a potential cod or salmon production, making an extended analysis of each. Technically, a modulated temperature strategy is tested at an experimental scale. However, further investigations should be developed before proposing any commercial scale initiative regarding to the balance required between the working strategy, fish species reared and seawater temperature profile. In fact, improving production’s technical efficiency both cost and environmental impacts could be reduced mostly due to a decrease in the energy use. On the other hand, a widespread analysis is made about the energy use and the main issues whiles the management of RAS. The importance of this technology worldwide and consumer’s consciousness towards environment’s welfare makes necessary to improve every aspect of RAS to increase the number of companies pointing to a more sustainable fish industry.

Future perspectives

The aquaculture sector continues to expand in a world where water, land, and fishery resources are under pressure to meet multiple human demands. Moreover, concerns regarding the safety of seafood products will likely remain or increase in the future, with consumers demanding seafood that can be guaranteed safe. RAS operations strive to control all aspects of production and can therefore remove or treat contaminants most effectively. RASs are able to guarantee reduced environmental impacts. All wastes can be concentrated and treated or used as an input to other production systems (e.g. agricultural fertilizer or methane generation). At the same time, RASs can be built in biosecure facilities away from water bodies. Such technology serves as a favorable technological fix although it rarely works well economically, especially for large-scale commercial systems. RAS are expensive, usually require costly indoor spaces and have continuous and substantial operational requirements beyond traditional methods. Moreover, as shown in this study, the costs of labor and energy can be prohibitively high depending on the location. In fact, RAS are likely to be hampered by rising electricity generation and fuel costs. Therefore, innovations in new energy sources (i.e. renewable sources) and energy saving measures could help alleviate the energy constraint.

Production location influences the competitiveness of the aquaculture farm by conditioning environmental impacts and economic expenditures factors. For instance, any comparison between different RAS productions will not be realistic as many factors (e.g. species, country, production volumes and policies) affect the results obtained. Therefore, although it will be challenging to design the most innovative, productive, efficient, profitable and environmentally friendly RAS in the future, it will not be a single measure that will lead to the widespread success of commercial RAS. Rather, the commercial success of this industry will require the need to focus on the combination of all aspects of these types of ventures. In reality, rethinking such productions with an integrated mind-set will help tackling holistically the simultaneous challenges of energy demands, management matters, consumers' preferences, and system's efficiency.

From governments' perspective, it more funding would benefit RAS researchers to help them provide consumers with more sustainable and safer seafood aquaculture products:

- Incentiving "greener production" by implementing renewable energies as main energy sources.
- Increasing production or marketing tariffs to non-sustainable productions.
- Promoting aquaculture products making alliances with supermarkets and the industry.

In such manner, the RAS manager of a future farm will give importance to the production's sustainability. In case of an ongoing farm, energy saving measures should be implemented by the adoption of energy audits as the main methodology.

Increase aquaculture awareness: informative courses at different educational levels (from early ages to university students) would promote the motivation and knowledge about the aquaculture industry.

From consumers’ perspectives, more efforts should be done educating consumers about the benefits of the products produced in RAS which offer both short and long period of time, and the values of local and sustainable products.

Conclusions and Thesis

Taking into account the objectives of this thesis and the questions that it aimed to answer, we can conclude that:

1. Basque coast local seawater temperatures influenced the growth performance of cod and salmon individuals. Both species experienced a halt in growth during warmer water temperature periods, but they were able to adapt to feast-and-famine conditions by showing marked growth spurts when environmental conditions, and food supplies were increased after a period of starvation.

2. Locally grown cod and salmon nutritional profiles showed acceptable protein, ash, humidity and lipid contents. These aspects were affected by the rearing conditions (i.e. temperature), but the marketable products were as good and of similar quality as other products (i.e. wild and different origin individuals).

3. Locally grown cod and salmon were equally accepted by the consumers when compared with wild caught and/or other farmed individuals (i.e. different origin and farming method). In both cases, the organoleptic tasting revealed that the quality of locally reared fish was equal or of similar characteristics to the samples obtained from the fish market. In case of cod, seafood experts were not able to discriminate between samples or origins (i.e. aquaculture and wild), while in the case of salmon the purchasing intention resulted to be more likely towards the locally grown fish than the commercial product. This shows the positive acceptance that consumers would have in relation to a possible local product.

4. The economic study undertaken concluded that land-based scenarios producing less than 200 t of cod fish may not be economically viable in a geographic zone where both salary and energy costs are limiting factors. Minimum marketable product’s price required to remain profitable was too high for the current market. Moreover, results showed that pilot-scale research models are costly and risky, leading to the general acceptance that their main role is to improve foundational understandings and contribute to applied scientific knowledge. Therefore, a clear dimension and perspective of economies of scale should be considered if affordable operational costs and consistent marketable final product prices are intended in the region, looking for a balance between operational costs and production benefits.

5. RAS are environmentally sustainable fish farming technologies although their high energy use is still an economic constraint in certain production areas such in those with high water temperature gradient profiles. Every energy flow within the system should be continuously monitored in order to optimize their efficiency. Being aware of the factors influencing the fluctuation of energy consumption would aid in implementing energy saving measures and thus result in operational cost reductions. An energy audit should be implemented at commercial scale RAS farms in order to have an economic and environmental sustainable fish farming system.

6. Fossil fuel based on-farm electricity for the on-growing of fish was shown to be the most environmentally unfriendly input. Hence, non-renewable energies are still the main energy sources used in the industry. Thus, renewable energies should be promoted in RAS as long as they are placed in suitable locations with access to energy sources such as solar, wave, hydro, thermo-solar, and domestic hot water. Additionally, cogeneration technique with biomass or other types of clean fuels should also be evaluated.

7. RAS management and understanding affects their performance and consequently the industry's success. Moreover, the investment required and the long pay-back periods were the greatest constraints. Thus, a good market or social study is required in order to plan an affordable and realistic production goal. In fact, the first requirement would be a reliable operation followed by low operating costs. Additionally, more communication between universities, researches and the industry is needed in order to train functional skills to future RAS managers. At the same time, sharing experiences and issues among different parties would be beneficial for industry's development. Rethinking RAS production with an integrated mind-set is needed to tackle the simultaneous challenges of energy demands; management matters; consumers' preferences; and system's efficiency.

8. The combination of Life Cycle Assessment and on-farm energy audit represents a useful tool to assist operators to eco-design or optimize their production. Likewise, it increases the speed and transparency of governance and decision-making, taking into account the time-based fluctuation of the energy consumption throughout the production cycle.

Thesis

"The farming of cod and salmon as cold-water species in a technically and socially sustainable way using the local seawater's temperature profile is feasible using Recirculating Aquaculture Systems (RAS) in the Basque coastal area (Northern Spain). Nevertheless, in order to be environmentally and economically sustainable new energy sources (i.e. renewable energies) should be tested and/or system's efficiency should be improved by implementing energy audits and/or time-based energy and resource saving measures as a way to test technologies viability".

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Zohar, Y., Y. Tal, H. J. Schreier, C. Steven, J. Stubblefield and A. Place. (2005). Commercially feasible urban recirculated aquaculture: Addressing the marine sector. Urban Aquaculture. B. Costa-Pierce. Cambridge, MA, CABI Publishing**:** 159-171.

Curriculum Vitae

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Home address: Aldapabarrena 38 Atico A **Town:** Leioa (Bizkaia)
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Education skills

2012- 2017 PhD on "Land based on-growing of Atlantic cod (*Gadus morhua*) and salmon (*Salmo salar)* using Recirculation Aquaculture Systems in the Basque Country: contributions to scientific understanding of economic feasibility, environmental sustainability, and societal acceptability"

2010-2011 MSc in Aquaculture Systems. University of Stirling – Institute of Aquaculture (Scotland)

2007-2010 MEng - Agronomy Engineering. Universitat de Lleida (Lleida –Spain)

2003-2007 BEng - Technical Agronomy Engineering. Navarra´s Public University (Navarra - Spain)

Language skills

Basque: native language
Spanish: 2nd native language
English: certificate in Advanced English (CAE) (equalisation to TOEFL 80-120/213-247/550-597)
Catalan: medium level
Czech: basic level

Additional skills

High level knowledge on SimaPro, AutoCad, Windows (Excel, Word, PowerPoint), Internet.
Medium level knowledge on IDRISI 32
Basic knowledege on SPSS, SAS, V-Fort, HyperLindo

Additional experience and personal merits/awards

2015- onwards National Students Coordinator at the European Aquaculture Society (EAS)

2012- 2016 Pilot-scale RAS modules management in a R&D facilitate (Northern Spain)

2016 September Student helper and National (Spanish) coordinator at the Aquaculture Europe 2016 (Edinburgh)

2015 October Student helper and National (Spanish) coordinator at the Aquaculture Europe 2015 (Rotterdam)

2014 October – Student coordinator at the Aquaculture Europe congress. Donostia-San Sebastián, 14-17 October.

2014 February – 2015 February: External researcher for the SeaFood Watch program of the Monterey Bay Aquarium (California)

2013 September - Recognition for being within the top 5 of the most downloaded papers in 2013 in *Aquaculture Engineering*. *"*Badiola, M., Mendiola, D., Bostock, J. (2012). Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges. *Aquacultural Engineering.* 51, 26-35"

2010-2011 MSc class delegate – University of Stirling

2007 *Best curriculum award*- Navarra Public University- Technical agronomy engineering

2005-2006 Erasmus delegate, Mendel University of Brno

2006-2007 Laboratory and lecturer assistant, Navarra´s Public University.

2006 European Inseminator title, Mendel University of Brno

2004-2005 Class-delegate at 2nd stage of the BEng, Navarra´s Public University

Grants

2015: Basque Government´s 3 months (1st June-28th August) stay permission to complete part of the PhD chapters in the University of Davis (California) supervised by Prof. Raul Piedrahita.

2013: Short period abroad studies Grant of the Basque Government – University of Davis- collaborating with Prof. Raul Piedrahita (July-October).

2012 PhD scholarship awarded by the Basque Government – AZTI Tecnalia Marine Research Division. PhD title: "Development of feasibility studies and recirculating technologies to produce cold water species (cod and salmon) in the Basque Country".

2010-2011 Basque Government grant for specialisation studies in a foreign country – Grant was used to do the MSc in Aquaculture Systems at University of Stirling, Institute of Aquaculture.

2010 Research Scholarship of the Basque Government to work within AZTI, for the development of aquaculture-based technological projects.

2005-2006 Erasmus grant, Mendel University of Brno (Czech Republic).

Courses and workshops

"Introduction to Generalized Linear Models with R"- Bilbao (Spain) November 2014

2nd Symposium of the Spanish network of Life Cycle Assessment (LCA). Derio (Spain) 6th November 2014

19th Annual Recirculating Aquaculture Systems Short Course (RAS, Aquaponics and Hydroponics technology) – Cornell University – July 2013

Social Life Cycle Assessment (SLCA) course. University of Pompeu Fabra (Barcelona). Duration of one week, given by International Life Cycle Academy.

Simapro ® software Advanced course (Life Cycle Assessment analysis). 3 days

Design of experiments Workshop. Faculty of Science and Technology, University of the Basque Country (UPV-EHU). 18 hours duration. December 2012

EAFE Workshop 2012, Green and Blue Growth: Challenges and opportunities for fisheries, aquaculture and the seafood industry. Bilbao 2-3 May 2012

RAS pilot scale module management course – IRTA Research Institution (Sant Carles de la Rápita, Spain) – January 2012

Sustainable Aquaculture summer course (Segovia (Spain), 2010; Laredo (Spain), 2012)

Presentations

Badiola, M. (2016). R&D+i towards the Sustainability and eco-desiging of Recirculating Aquaculture Systems (RAS). Plataforma Tecnológica Europea de Pesca y Acuicultura. Invited speaker. Madrid (Spain) 10th October.

Badiola, M., Basurko, O., Gartzia, I., Mendiola, D. (2015). Marketable size salmon (*Salmo salar*) production in the Basque region: a feasibility case study from a

Recirculating Aquaculture System (RAS). Aquaculture Europe 2015, Rotterdam (Holland). Conference paper. 20-23 October

Badiola, M., Albaum, B., Mendiola, D. (2015). A sustainability evaluation, based on environmental indicators of Recirculating Aquaculture Systems (RAS) applied to all countries and all species. Aquaculture Europe, Rotterdam (Holland). Invited speaker from Biomar. Conference paper. 20-23 October.

Badiola, M., Basurko, O., Gabiña, G., Mendiola, D. (2015). A combination of environmental assessment methods and key economic factors to improve RAS's sustainability. Aquaculture Europe 2015, Rotterdam (Holland). Invited speaker from Biomar. Conference paper. 20-23 October

Badiola, M., Albaum, B., Mendiola, D. "A sustainability evaluation, based on environmental indicators, of Recirculating Aquaculture Systems (RAS) applied to all countries and all species". NordicRAS 2015. Molde (Norway). September 29th-October 2nd 2015

Badiola, M., Cabezas, O., Gabiña, G., Mendiola, D. (2015). "Combination of Life Cycle Assessment and energy audit to reduce the environmental impacts of rearing cod in a pilot scale Recirculating Aquaculture System". LCM 2015. Bourdeax (France). Poster. 31st August- 3 September (2015)

Badiola, M., Cabezas,O., Curtin, R., García,M., Gartzia, I., and Mendiola, D. "Estudios de viabilidad sobre el crecimiento y produccion del bacalao (G*adus morhua*) hasta talla comercial". Dia de la Acuicultura. Invited speaker. PIE-Plentzia (Spain). November 28th 2014.

Badiola, M., Cabezas,O., Curtin, R., García,M., Gartzia, I., and Mendiola, D. "Land based on-growing of Atlantic cod (*Gadus morhua*) to marketable size – a feasibility study from the Basque Country (Northern Spain)". Aquaculture Europe 2014. Donostia-San Sebatián (Spain) 14-17 October.

Badiola, M., Cabezas, O., Gabiña, G., Mendiola, D. "Energy audits in Recirculating Aquaculture Systems (RAS): as a way forward to guarantee sustainability" (POSTER). Aquaculture Europe 2014. Donostia-San Sebatián (Spain) 14-17 October.

Badiola, M. and Mendiola, D. (2013). "Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges". Workshop - Finfish nutrition and aquaculture technology at the crossroads. Invited speaker. Bremenhaven (Germany). 18th February.

Badiola, M. (2013). Development of feasibility studies and recirculating technologies to produce cold water species (cod and salmon) in the Basque Country. RIMER 2013. Invited speaker. Donostia-San Sebastian (Spain). 8th February

Publications

Badiola, M., Piedrahita, R., Hundley, P., Basurko, O.C. Energy use in Recirculating Aquaculture Systems: A review. *Environmental Science and Technology* (submitted).

Badiola, M., Basurko, O.C., Gabiña, G., Mendiola, D. Integration of energy audits in Life Cycle Assessment (LCA) methodology to improve the environmental performance assessment of Recirculating Aquaculture Systems (RAS). *Journal of Cleaner Production* (accepted).

Badiola, M., Gartzia, I., Basurko, O.C., Mendiola, D. (2017). Land-based growth and sensory evaluation of Atlantic salmon (Salmo salar): assessing consumers acceptance. *Aquaculture Research* (accepted for publication January 1st 2017).

Badiola, M., Albaum, B., Mendiola, D. (2016). Land based on-growing of Atlantic cod (*Gadus morhua*) using Recirculating Aquaculture System (RAS); a case study from the Basque region (northern Spain). *Aquaculture*, 468, 428-441.

Badiola, M., Mendiola, D., Bostock, J. (2012). Recirculation Aquaculture Systems (RAS) analysis: Main issues on management and future challenges. *Aquacultural Engineering.* 51, 26-35.

Badiola, M. (2010). Construction and Design of a winery located in La Rioja (Spain) producing 400,000 litters of bottled and caned wine and sangria. MEng final project, Faculty of Agronomy, University of Lleida, 750p.

Badiola, M. (2007). Construction and Design of a winery located in Navarra (Spain) producing 225,000 litters of bottled and caned wine". BEng final project, Faculty of Agronomy, Navarra´s Public University, 1120p.

Technical Publications:

Badiola, M., Albaum, B., Mendiola, D. (2014). Recirculating Aquaculture Systems – Global – All Species. Seafood Watch program, Monterey bay Aquarium. http://www.seafoodwatch.org/-/m/sfw/pdf/reports/mba_seafoodwatch_global_ras_report.pdf

Hatchery International – Calculating sustainability for RAS: there is still work to be done. March/April 2015, page 41

Hatchery International – Can codfish be grown profitably in RAS? A study from Northern Spain says yes…but! September/October 2015, page 34-35.