Safeguarding the welfare of Scottish farmed Atlantic salmon: current practices and future prospects

Thesis submitted for the degree of doctor of philosophy

Institute of aquaculture

University of Stirling

July 2023

by

Timothy Robert Wiese

Declaration

This thesis has been composed in its entirety by the candidate. Except where specifically acknowledged, the work described in this thesis has been conducted by me, and has not been submitted for any other degree or qualification.

CANDIDATE NAME: Timothy Robert Wiese

SIGNED:

DATE: 15/07/2023

SUPERVISOR NAME: Sonia Rey Planellas

SIGNED:

DATE:29/01/2024

Acknowledgements

I would like to express my sincerest gratitude to Professor Jimmy Turnbull and Associate Professor Sonia Rey-Planellas at the University of Stirling, Dr. Susan Jarvis at the University of Edinburgh, and Professor Marie Haskell at Scotland's Rural College (SRUC). I could not have asked for a more exceptional team of supervisors, each of whom have provided me with continuous support and guidance throughout my entire PhD study. Thank you for sharing your wealth of experience and knowledge with me, for teaching me how to work, think, and write like a scientist, and above all thank you for providing me with this incredible opportunity to carry out this work. Thank you to everyone from the Scottish salmon farming sector who contributed their precious time and opinions while participating in the Industry survey and QBA sessions – these chapters would not have been as relevant to the industry or even possible without your help. A big thanks also goes to Malcolm Johnstone and the RSPCA Assured for providing the farm site compliance data along with many valuable insights into this industry. Thank you to Chessor Matthew, Pedro Munoz, and David Bassett from the Marine Environmental Research Laboratory in Machrihanish for your support in the QBA experiment.

Significant accomplishments are rarely achieved without the support of family and friends. To Wesley, Callum, Christian, Judith and Ana, thank you all for being there for me through thick and thin. To my parents, I could write an entire page acknowledging what you have done for me. I am so incredibly grateful for the love, guidance, and opportunities that you both have given me. The same goes for my big brother, who has been my role model for many years now and has consistently pushed me to becoming a better man. Thank you for dropping your occasional wisdom bombs, and for helping me to keep calm when I needed to the most.

To the love of my life and soon to be mother of my child, MT, a thank you is not enough. You have brought me infinite joy and been my rock ever since you came into my life. Without you, I would not be where I am today and I cannot imagine a future without your love and support.

Finally, a thank you to my son who I will be meeting very soon. Thank you for igniting a fire within me and fuelling my work ethic to new heights this year. You are already shaping who I am becoming as a man, and given me a new purpose in life. I cannot wait to see what little experiments you do of your own when you finally come into this world.

Abstract

Farmed fish welfare has become a growing priority as aquaculture continues to expand to meet global demand. The Scottish salmon farming industry is a prime example of this growth, reaching record highs in production and intensification over recent years. The compelling evidence for fish sentience highlights the ethical imperative of safeguarding the welfare of the millions of animals involved. Achieving appropriate levels of salmon welfare, however, presents considerable challenges. Animal welfare is a complex, multi-faceted concept, the intricacy of which is only further amplified when dealing with the anadromous life cycle of Atlantic salmon. The aim of this PhD was to provide industry-relevant contributions towards the monitoring and safeguarding of farmed salmon welfare. An additional aim was to validate or further refine a novel on-farm welfare assessment tool that provides the most benefits in this manner. Chapter 1 provides the context for this study, outlining key concepts of animal welfare, the importance of farmed salmon welfare, and various factors, indicators, and considerations that are important for farmed salmon welfare. Chapter 2 addresses the complexity of enhancing farmed salmon welfare by conducting a survey on the Scottish salmon farming sector, consulting industry professionals to better understand their current welfare concerns and research priorities. Chapter 3 investigates what role welfare standards can play in providing assurances for farmed salmon welfare, as well as how welfare practices within the industry have changed over the years, through examining changes in farm site compliance to these standards. Chapter 4 assessed the effectiveness of Qualitative Behavioural Assessment (QBA) in capturing changes in the behavioural expressions of Atlantic salmon following exposure to a stressful event. Chapter 5 summarises the findings from these studies, outlining how Chapters 2 and 3 informed the development of the QBA experiment conducted in Chapter 4 and the significance of QBA's validation. Chapter 5 then develops on these findings, proposing a direction for future research regarding the potential for behavioural welfare assessment tools to utilise computer vision and machine learning technologies. The results from this thesis highlight the potential that non-intrusive, remote, animal-based welfare indicators have in improving the monitoring and management of farmed salmon welfare. In particular, QBA shows great potential as a unique welfare indicator within aquaculture. This is the first study to demonstrate QBA's sensitivity to changes in the behavioural expressions of Atlantic salmon and highlight the unique insights it offers into salmon welfare.

Conferences & Presentations

<u>Oral Presentations</u>

CPD course (Institute of Aquaculture) **Stirling, UK, June 2023**

Titled: ‘Scottish salmon & RSPCA Assured – welfare standards’.

Stirling PhD conference **Stirling, UK, October, 2022**

Titled: ‘Concerns and research priorities for the welfare of Scottish farmed Atlantic salmon (*Salmo salar*) – An industry perspective’.

European Aquaculture Society (EAS) **Rimini, Italy, September, 2022**

Titled: ‘Scottish farmed Atlantic salmon welfare (*Salmo salar*) – An industry perspective’.

Seminar (Institute of Aquaculture) **Stirling, UK, July, 2021**

Titled: ‘Developing on-farm welfare assessments of Atlantic salmon; industry appraisal & upcoming experimental chapters’.

Seminar (Roslin Institute – Animal behaviour & welfare) **Edinburgh, UK, July, 2020**

Titled: ‘Welfare monitoring and assessment of farmed Atlantic salmon’.

CPD course (Institute of Aquaculture) **Stirling, UK, February 2020**

Titled: ‘Welfare: what is it, and how to assess it?’.

<u>Poster Presentations</u>

Stirling PhD conference **Stirling, UK, October, 2022**

Titled: ‘Concerns and research priorities for the welfare of Scottish farmed Atlantic salmon (*Salmo salar*) – An industry perspective’.

Publications

Chapter 2 – 'Concerns and research priorities for Scottish farmed salmon welfare – An industry perspective'.

A close derivative of this work has been published as:

Wiese, T. R. et al. (2023) 'Concerns and research priorities for Scottish farmed salmon welfare – An industry perspective', Aquaculture. Elsevier B.V., 566(July 2022), p. 739235. doi: 10.1016/j.aquaculture.2023.739235.

Contributions:

Timothy Robert Wiese: Conceptualisation, visualisation, methodology, data curation, writing – original draft, writing – review & editing, investigation, formal analysis. **Marie Haskell:** supervision, conceptualisation, visualisation, methodology, writing – review & editing, validation. **Susan Jarvis:** Supervision, conceptualisation, visualisation, methodology, writing – review & editing, validation. **Sonia Rey-Planellas:** Supervision, conceptualisation, visualisation, methodology, writing – review & editing, validation. **Jimmy Turnbull:** Supervision, conceptualisation, visualisation, methodology, writing – review & editing, validation.

Chapter 3 – 'Farmed salmon welfare practices – insights gained from evaluating standards and farm compliance'

A close derivative of this work will be submitted for publication.

Contributions:

Timothy Robert Wiese: Conceptualisation, visualisation, methodology, data curation and formal analysis, writing – original draft, review & editing, investigation. **Susan Jarvis:** Supervision, conceptualisation, visualisation, methodology, writing - review & editing, validation. **Marie Haskell:** Supervision, conceptualisation, visualisation, methodology, writing - review & editing, validation. **Sonia Rey-Planellas:** Supervision, conceptualisation, visualisation, methodology, writing - review & editing, validation. **Jimmy Turnbull:** Supervision, conceptualisation, visualisation, methodology, writing - review & editing, validation.

Chapter 4 – 'Application of Qualitative Behavioural Assessment (QBA) as a welfare indicator for farmed Atlantic salmon (*Salmo salar*) in response to a stressful challenge'

A close derivative of this work has been published as:

Wiese, T. R. et al. (2023) 'Qualitative Behavioural Assessment (QBA) as a welfare indicator for farmed Atlantic salmon (*Salmo salar*) in response to a stressful challenge', Frontiers in Veterinary Science – Animal Behaviour and Welfare. Elsevier B.V., 10 (Sep 2023). Doi: https://doi.org/10.3389/fvets.2023.1260090.

Contributions:

Timothy Robert Wiese: Conceptualisation, visualisation, methodology (recording experiment – Machrihanish, QBA process), recording, editing, and preparing of footage used in study, data wrangling and statistical analysis, writing – original draft, writing – review & editing, investigation. **Sonia Rey-Planellas:** Supervision, conceptualisation, visualisation, methodology (recording experiment – Machrihanish), writing & statistical analysis – review & editing, validation. **Marie Haskell:** Supervision, conceptualisation, visualisation, methodology (recording experiment – Machrihanish, QBA process), writing & statistical analysis – review & editing, validation. **Susan Jarvis:** Supervision, conceptualisation, visualisation, methodology (recording experiment – Machrihanish, QBA process), writing & statistical analysis – review & editing, validation. **Andrew Davies:** Recruitment of participants for term generation and QBA scoring, Writing – review & editing, validation. **Francoise Wemelsfelder:** Methodology (QBA process), QBA training, recruitment of participants for term generation and QBA scoring, writing – review & editing, validation. **Jimmy Turnbull:** Supervision, conceptualisation, visualisation, methodology (recording experiment – Machrihanish, QBA process), writing & statistical analysis – review & editing, validation.

Table of Contents

Figure list

Table list

CHAPTER 1. General introduction and literature review

1.1 Animal welfare – background and general concepts

With aquaculture continuing to expand to meet the demands of a growing global population, fish welfare has now cultivated a place amongst the industry's list of priorities. Animal welfare science is a broad topic, encompassing everything that affects the physical and emotional state of an animal, its ability to cope with the surrounding environment, and its overall quality of life (Webster, 2016). How welfare is defined will largely determine how it is assessed and, consequently, what information is obtained from subsequent studies (Fraser *et al.*, 1997; Huntingford and Kadri, 2009). One of the challenges in defining a concept like welfare is keeping it within the realms of which it can be objectively evaluated (Ashley, 2007; Dawkins, 2015). This is because ideas regarding consciousness are often central to welfare (Dawkins, 1990, 1998, 2015; Fraser *et al.*, 1997), and the subjective nature of a conscious experience is combined with our bewilderment as to how these experiences arise from brain tissue in the first place (Dawkins, 2015). As a result, direct measurements of individual experiences cannot yet be made if we are to maintain an objective standard. If animal welfare is to be studied empirically, we must then take this problem into account and look for the closest correlates of consciousness (Dawkins, 2015). We can then attempt to use other information from the animal, such as physiological or behavioural data, to make links and validated inferences from their functional states in determining how the subjective experiences of animals are likely to be influenced (Fraser *et al.*, 1997; Mellor, 2012).

Mistaken notions about the concept of welfare have often led to difficulties in defining and assessing welfare (Dawkins, 1998). Instead of expecting welfare to be this single, quantitative expression that is always valid, Dawkins advocates that welfare should be regarded "as a trait having multiple attributes and being different under various circumstances" (Dawkins, 1998, p. 307). In this approach, welfare is equated to assessing the safety rating of a building, rather than measuring the height. Accordingly, no single welfare measure is considered reliable enough in isolation (Ashley, 2007). There are instead a variety of "measures", or indicators, all of which need to be considered together while assessing this multi-faceted concept. In determining which indicators are important for welfare, three different but overlapping views have been established (Fraser *et al.*, 1997; Fraser, 2003):

1) A functions-based approach: Focusing on the animal's ability to physically adapt to its current environment with normal functioning of physiological and behavioural systems, allowing for sufficient health and growth.

2) A feelings-based approach: Focusing on the subjective mental state, where animals should be free from negative experiences such as prolonged and intense fear or pain while also experiencing pleasures.

3) A nature-based approach: Focusing on the ability of an animal to lead a natural life and express their inherent biological nature through the development and use of their species-specific adaptations.

1.2 Arguments between the approaches

1.2.1 On a function-based approach

The concerns of an animal's welfare are often closely related to its overall condition, including directly measurable variables such as disease, injury, or death (Broom, 1991; Dawkins, 1998). Measuring an animal's health as a correlate to its welfare also avoids the dilemma of measuring the subjective experience of an animal (Dawkins, 2015).

There is the argument that biological functioning itself is important, regardless of how it may affect an animal's subjective experiences (Fraser *et al.*, 1997). Strictly following this reasoning, the well-being of an animal depends not on what it prefers, but also on the things that benefit the animal whether or not the animal is consciously aware or in pursuit of them (Broom, 1991). Broom provides such examples of how the welfare of an individual could be considered negatively impacted without any suffering occurring:

1) Injuries without feeling any pain due to endogenous analgesic opioids (or artificial analgesics); the argument here is that the state of the animal is affected, and the injury itself is an indicator of poor welfare.

2) Difficult housing conditions could impair an animal's immune system function and increase susceptibility to disease. Even without disease developing, one could argue that the state of the animal is directly affected and is thus an indicator of poor welfare.

3) Housing conditions could involve sensory deprivation to the point where the animal develops minimal "normal" behaviour, which the animal copes with by self-narcotizing. The endogenous opioids may mean the animal is not suffering, but the modification of its own state to cope with the conditions could again be seen as an indicator of poor welfare.

The concept of welfare used in these examples encompasses concerns that go beyond suffering, and puts additional value on the state of the individual regardless of how it perceives its own welfare. Determining whether such an approach is valid or not becomes more of a debate of ethics and morality, rather than science. Many argue that, in the case of animal welfare, it would be best to apply the precautionary principle (Jones, 2013; FAWC, 2014; Sneddon, 2015). This involves situations where there is an issue of potential harm (in this case, an impaired functional state), and the significance of this cannot be determined by scientific knowledge alone. When applying the precautionary approach in this case, the importance of the potential harm is assumed, and care should be taken in preventing this from occurring.

In conceptualising welfare solely in terms of biological functioning, there is no obvious point in the continuum of bodily responses where welfare can be determined as impaired (Fraser *et al.*, 1997). There is no way of calibrating what threshold of a physiological variable, in isolation, is compatible with poor or good welfare (e.g., changes in hormones, corticosteroid levels, neurotransmitters) without evidence directly relating the variable to the animal's health or behaviours of aversion / attraction (Dawkins, 1998).

Another issue with a strictly function-based approach is that animals can be in good health, but still suffer when prevented from performing certain behaviours (even when there is little to no risk to fitness) (Dawkins, 1990). Dawkins argues that, if an animal perceives itself to be in great danger when it cannot perform certain behaviours, then it will suffer even if it is not actually in danger.

1.2.2 On a feelings-based approach

The subjective experience of an animal is arguably of ultimate concern in animal welfare (Dawkins, 1990, 1998, 2015; Fraser *et al.*, 1997). Certain symptoms of poor welfare that fall under this category, such as pain and fear, are suggested to have evolved in many animals as a defence mechanism against threats to survival (Dawkins, 1998). These 'defences' are unpleasant by design; pain evolved because, by being unpleasant, it keeps an animal away from the larger "evolutionary disaster" of death. Fear evolved to come into play before physical injury occurs; pain helps avoid death, fear helps avoid pain and/or death (Dawkins, 1998). Natural selection produced such mechanisms to act on the animal at earlier and earlier stages, long before their health and fitness is in danger. Since emotional states have such a fitness value (Korte, Olivier and Koolhaas, 2007), it is reasonable to assume that many animals have evolved such traits. It is therefore important to consider welfare measures outside of the obvious ones of disease and injury (Dawkins, 1998).

An important consideration with unpleasant states is determining the point at which these are deemed to negatively impact the animal's overall welfare. While pain, fear, hunger, boredom, and other relevant states can obviously have negative effects on welfare, there is no justification for deciding that such affective states always compromise an animal's welfare when they are "an unavoidable part of normal animal life" (Huntingford *et al.*, 2006, p.335).

To be completely free of these unpleasant states is idealistic (Korte, Olivier and Koolhaas, 2007; Green and Mellor, 2011). One should attempt to decide at what point the frequency or severity of these unpleasant states significantly impact an animal's overall welfare; i.e., when an animal is suffering. Dawkins (1990) suggests that suffering occurs when there is an acute or prolonged experience of an unpleasant mental state because the animal is unable to respond in a way that would normally reduce risks to life and reproduction in those circumstances. The 'Allostasis' model, introduced into the animal welfare discussion by Korte et al. (2007) falls into this more dynamic view of considering the animal's welfare (Figure 1-1).

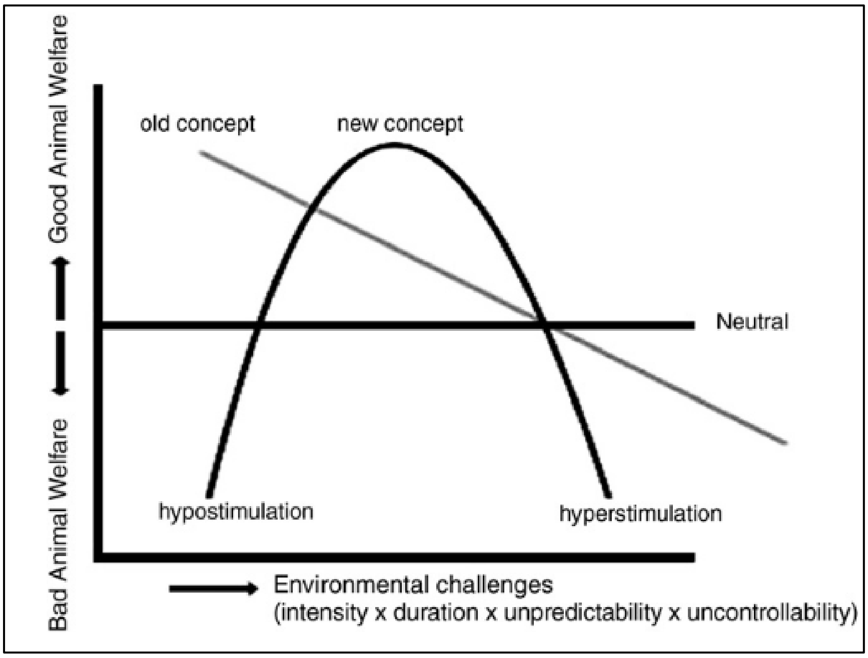

Figure 1-1. Comparison of animal welfare concepts in relation to increasing environmental challenges. The straight line is based on older concept of homeostasis, whereas the inverted U-curve is based on the concept of allostasis. Source: (Korte, Olivier and Koolhaas, 2007).

Rather than focusing on maintaining the animal's physiological, behavioural, and emotional affective states within this constant range that is accepted as some stable equilibrium (i.e., maintaining 'homeostasis'), the ability for the animal to change their coping strategies in anticipation to incoming (along with currently existing) challenges / stressors is what matters the most (Korte, Olivier and Koolhaas, 2007; Green and Mellor, 2011). Therefore, the primary concern should be avoiding situations where the challenges presented are too much, or perhaps too little, for the animal to cope with.

Limiting welfare to a solely feelings-based approach presents its own issues. There is first the problem of gaining empirical information on the subjective experience of an animal. This approach is also solely dependent on the animal having conscious experiences in the first place (Huntingford *et al.*, 2006). In addition, focusing solely on emotional state disregards how closely linked health is to welfare. In the example of a habitual smoker, smoking cigarettes causes pleasure and relieves distress; the lung damage causes little immediate suffering, and there are no grounds to determine whether the future suffering outweighs the pleasure (Fraser *et al.*, 1997). A strictly feelings-based approach could possibly lead to the conclusion that, in some cases, cigarettes have a net improvement in welfare, even though they drastically impair health.

1.2.3 On a nature-based approach

Closely related to the feelings-based approach, this concept brings forward the argument of how an animal's welfare can be compromised even in perfect health, and that "what is natural is inherently good" (Huntingford *et al.*, 2006, p.334). However, the cost felt by an animal when deprived of 'natural' conditions may differ depending on the type of neglect that has occurred.

Preventing a certain behaviour that has little consequence to the animal's fitness may have a different cost to the animal than artificially depriving it of commodities and / or preventing it from performing innate behaviours (Dawkins, 1990). Dawkins (1990) provides an example of a bird that normally migrates being kept in a cage. Although it may be well cared for, and its chances of survival are far higher than its wild counterparts, the bird is unable to fulfil its strong motivations to fly. Since the bird has not evolved to meet its new conditions, this may mean that the bird's welfare is compromised regardless of its physical health. This example could be extended to any other species where innate behaviours may be restricted due to husbandry conditions.

Although the inherent biological nature of an animal is an important consideration for their welfare, living a natural life gives no guarantee that the full range of ethical concerns in the animal's welfare will be satisfied (Fraser *et al.*, 1997). Animals kept in entirely 'natural' environments may still suffer greatly if its adaptations are insufficient to meet the challenges they are exposed to, including severe temperatures outside of their thermoregulatory ranges (Fraser *et al.*, 1997). Aggressive competition and predation are also unavoidable parts of natural life for many species, which are arguably detrimental to their welfare. Additionally, there are major empirical and conceptual problems that arise when defining the 'nature' of a given animal (Fraser *et al.*, 1997). Therefore, although a nature-based concept is an important factor when considering the welfare of an animal, it cannot by itself provide sufficient guidelines for defining and assessing welfare.

1.3 Integrating the three approaches into one concept of welfare

As apparent in the examples given above, these three different approaches involve a considerable but incomplete overlap, and separately they can lead to conflicting conclusions on how an animal's welfare should be judged (Fraser *et al.*, 1997; Huntingford *et al.*, 2006; Fraser, 2008). Fraser (1997) makes an important point that, while science may be able to provide empirical information relevant to welfare, it cannot turn such conflicting confusions into purely empirical matters by choosing one conception of animal welfare to the exclusion of others. Fraser concludes that, if the research is to address the major ethical concerns regarding animal welfare, then the conception which scientists adopt must reflect the whole range of ethical concerns existing in society (Function, Feelings, and Nature).

In working towards improving animal welfare, an integrated approach will need to be utilised that considers the three different approaches together, balancing different benefits and seeking options to address all three views (Fraser, 2009). This approach has already been touched on in the first paragraph of this review. It is understood that affective experiences are not solely a reflection of the animal's functional state; they can also have a direct impact on the animal's functional state (Green and Mellor, 2011; Hemsworth *et al.*, 2015). The two approaches are intimately connected with each other (see Figure 1-2). Attempts can and should be made to use a combination of physiological and behavioural data to make as many validated inferences on the animal's subjective experience as possible.

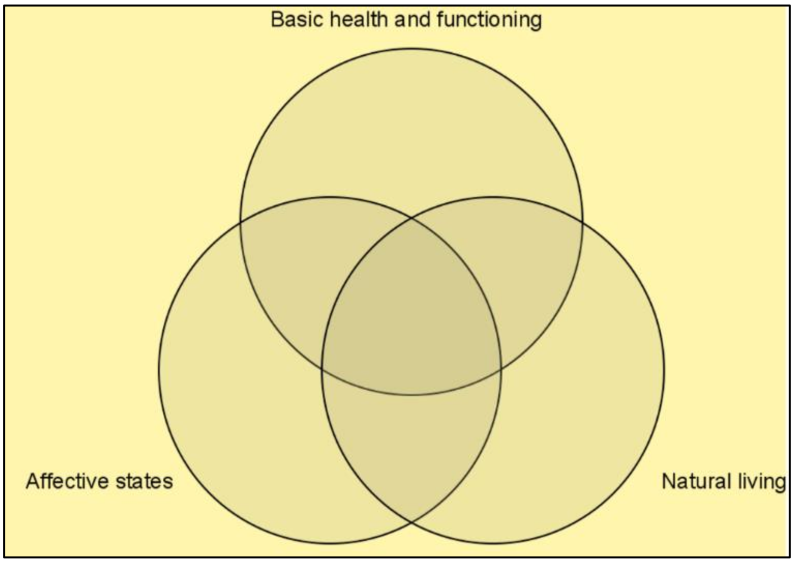

Figure 1-2. The three conceptions of animal welfare and their overlapping nature (Fraser, 2009).

In integrating these three concepts together, it could prove useful to recognise that there are three classes of problems which may arise when the adaptations possessed by an animal do not fully correspond to the challenges posed by its current environment (Fraser *et al.*, 1997). Situations where animals are placed in artificial conditions are often those which produce a degree of disconnection between the animal's natural behaviour, affective states, and basic health and functioning (Fraser *et al.*, 1997; Fraser, 2009). This disconnect is often responsible for the disconnect between these three concepts of welfare explored so far. Figure 1-3 below illustrates how these different approaches may be considered together.

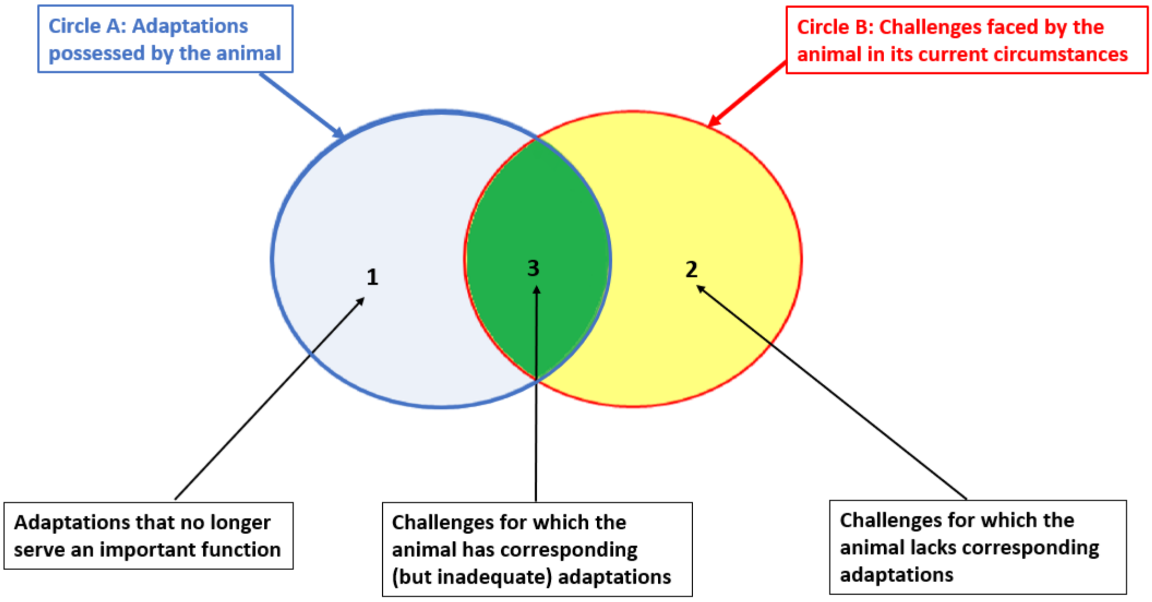

Figure 1-3. Personalised version of the conceptual model by Fraser et al. (1997), illustrating the three broad classes of problems that may arise when the adaptations possessed by the animal (Circle A) make an imperfect fit to the challenges it faces in the circumstances which it is kept (Circle B).

Circle A represents the adaptations of the animal, which can involve anatomy, physiology, or behaviour; this can also include affective states (hunger, cold, fear, pain etc.) that motivate the animal to act in certain ways. Circle B represents the challenges that the animal is facing in its current circumstances, which can include any challenging condition (exposure to cold temperatures, pathogens, predators, malnutrition, sensory deprivation etc.) (Fraser *et al.*, 1997).

Area 1 to the left of the overlap involves examples where animals possess adaptations that no longer serve a significant function in their current environment. Here, unpleasant subjective experiences may arise, yet these may not be accompanied by significant disruption to biological functioning, such as a bucket-fed calf experiencing a strong, frustrated desire to suckle even though it's well fed (Fraser *et al.*, 1997).

Area 2 to the right of the overlap involves examples where the environment poses challenges for which the animal has no corresponding adaptation. An example of this is how some species of fish fail to avoid certain contaminants (phenol, selenium) even at life-threatening levels. These problems may cause serious biological functioning impairment, yet the animals may show no accompanying effects on their subjective experience (Fraser *et al.*, 1997).

Area 3 in the central overlap area involves examples where the animal faces challenges for which the animal has corresponding but inadequate adaptations. For example, fluctuating ambient temperatures for which the animal's thermoregulatory adaptations are inappropriate. In this case, the animal's affective experience as well as its functional state are impacted (the animal both feels and functions poorly) (Fraser *et al.*, 1997). This model is one way of conceptualizing an integrated approach to the three ranging views of welfare concerns that must be addressed by animal welfare research.

From this, a working definition of welfare could be: ‘The state of an individual with regards to their physical condition and accumulation of positive and negative subjective experiences over time’. It is important here to recognise that giving a certain value to any aspects within this definition will inevitably involve some degree of subjectivity. Therefore, each aspect (physical condition, ‘positive’ experiences [i.e., pleasure], ‘negative’ experience [i.e., suffering]) should all be given equal weight when considering the welfare status of an animal.

1.4 Fish neurophysiology and behaviour – arguments for or against their ability to perceive their own welfare

For the purpose of this chapter, it is important that the working definitions for the following terms are clearly outlined:

Cognition: The broader mental action of acquiring, processing and transforming sensory data into information which the organism involved can then conceptualise.
Consciousness: An ‘umbrella’ term, used to **describe the varying degrees of awareness found within organisms.**

↓

Awareness: The ability of an entity to sense events, objects, or sensory patterns and respond to them. This, however, does not necessarily imply an understanding of the events themselves, or a capacity to feel, perceive, or experience on a subjective level.

↓

Sentience: A specific branch of consciousness that involves more complex, private, and subjective experiences. Rather than simply being ‘aware’ of one’s surroundings, **a sentient individual is also ‘aware’ of itself within the context of those surroundings**. **This ‘self-awareness’ allows an understanding behind one’s surroundings and what they mean to the individual themselves.** Consequently, this then includes the capacity of an individual to specifically feel, perceive, or experience on a subjective level (i.e., to experience pleasure or suffering)**.**

There is an ongoing debate that certain levels of consciousness, even to the point of sentience, are the sole prerequisite for the consideration of welfare in an animal (Dawkins, 1990, 2004; Chandroo, Duncan and Moccia, 2004; Broom, 2007). This view disregards that the general state of an animal may have any inherent value. Determining whether this exclusive approach is valid, or if an all-inclusive approach (both feelings and function matter) is more appropriate, becomes a matter of opinion rather than scientific debate. Nevertheless, if it can be determined that fish are sentient and therefore capable of perceiving their own welfare, then the welfare of fish is of our concern regardless of which approach is taken.

1.4.1 Inferring consciousness from complex behaviours

When the nervous system of a species attains a certain level of complexity during evolution, it is assumed that animals are able to develop ‘primary consciousness’, where they can form and act upon internal neural representations of their internal and external environment for the purpose of directing the animal’s behaviour (Chandroo, Duncan and Moccia, 2004).

Indirect evidence for such conscious, motivational affective states come from neuro-anatomy, neuro-physiology, and behaviour, with animal behaviour being one of the best windows into an animal’s subjective state (Chandroo, Duncan and Moccia, 2004). It is important to recognise, however, that behavioural changes can arise because an animal is either mentally aware of a situation, or because changes occur in the neurophysiology of the animal without any conscious awareness. For an animal to be able to be aware of its own circumstances, this may require some degree of cognitive sophistication (Braithwaite, Huntingford and Van den Bos, 2013).

This cognitive ability can be identified in animals by testing whether learned information is retained by an animal as procedural (reflexive response) or as declarative (flexible and adaptive response, i.e. consciously perceived) (Chandroo, Duncan and Moccia, 2004): Procedural representations involve simply reacting to a stimulus without awareness about the consequences of the response. Declarative representations involve selective attention to internal and external stimuli, anticipation, expectation and goal directed activity, therefore allowing for increasingly flexible behaviour and adaptive responses.

1.4.2 Fulfilling criteria for consciousness – evidence for declarative representations in fish

It has been argued that such declarative representations are required for consciousness to occur (Taylor, 2001; Chandroo, Duncan and Moccia, 2004). Studies have shown the ability of Siamese fighting fish to gather information on the relative fighting abilities of other conspecifics simply through the observation of aggressive interactions between other individuals (Oliveira, McGregor and Latruffe, 1998; McGregor, Peake and Lampe, 2001). The behavioural responses of the observer fish were sensitive not only to changes in the level of aggressiveness displayed by previewed fish, but also to the outcome of a conflict (McGregor, Peake and Lampe, 2001). Individual recognition and the ability to assess the fighting ability of future opponents in order to flexibly alter their fighting strategy, based solely on observation, is unlikely to be remembered as a procedural representation. Another example of this ability is the recognition of dominant aggressors in Atlantic salmon and body darkening as a submissive communication in order to avoid costly fights (O'Connor, Metcalfe and Taylor, 2000). To use observational learning through visual cues would suggest that these fish can retain memories as declarative representations (Chandroo, Duncan and Moccia, 2004).

In a study by Bass and Gerlai (2008), zebrafish that had not been previously exposed to predatory fish exhibited a significantly elevated fear response (increased number of jumps) to their natural predator (Indian Leaf Fish). This was not evoked by the sight of an allopatric predator (Compressed Cichlid). Such a difference in responses to two predatory fish of similar size and capabilities demonstrated that zebrafish were capable of selectively responding to different stimulus fish species without prior exposure to either kind. Although this does not provide direct evidence for consciousness in fish, it further fulfils one of the prerequisites for consciousness (having higher cognitive capabilities, i.e., declarative representations). Behavioural changes that may have a protective function in response to potentially damaging/painful events are also important indicators of a negative affective component (such as pain or fear) that could be associated with the sensory experience (Sneddon, 2009).

1.4.3 Nociception and pain perception in fishes

A major consideration in determining sentience within fish is on their ability to construct cognitive representations of noxious events (i.e., to feel pain). This is because pain is an affective experience which requires the animal to have conscious awareness (Rose, 2002). An important distinction to be made in understanding pain is its difference from nociception (Rose *et al.*, 2014). To experience pain, animals must respond to potentially painful events in a way that shows the animal is not simply exhibiting a nociceptive reflex (i.e. changes in the behaviour are not just a reflex response) (Sneddon, 2003; Sneddon, Braithwaite and Gentle, 2003a; Sneddon, 2015). Sneddon (2015) outlines two key criteria in establishing pain perception, which are characteristic of declarative representations:

1) A whole-animal response to painful events: nociception of a painful stimulus is conveyed to the CNS, where central processing occurs and innervates motivational/emotional behaviour and learning. Behavioural and physiological alterations occur outside of simple reflexes, with long-term responses including avoidance and protective behaviours. These reactions should also be reduced by the use of analgesics or painkillers.
2) The pain experience should influence the animal's future behavioural decisions on such an event, such as seeking analgesia or paying a cost to reduce its pain, or

avoiding the noxious stimulus and learning to avoid future encounters (adaptive responses).

In a study assessing the responses of trout to injections of noxious stimulants in the lip, fish exhibited profound differences in their behaviours between treatment groups (Sneddon, Braithwaite and Gentle, 2003a). Fish injected with a venom or acid took more than double the time to begin ingesting food again, in comparison to the control/saline injected fish. Those injected with venom or acid also performed anomalous behaviours, including ‘rocking’ and, specifically those in the acid group, rubbing of their lips into the gravel and tank walls. Performances of anomalous behaviours during painful events usually occur within the short time period where the pain is the most intense (Sneddon, Braithwaite and Gentle, 2003a). Coincidentally, the rocking behaviour performed by the trout was observed only in the 1.5 hours after injection. The researchers argue here that such behaviours as rocking and rubbing of the affected area are complex in their nature, and as such may not be simple reflexes. This study concluded that the behaviours of teleost fish can be adversely affected to the point where such behaviours strongly suggest a significant level of discomfort.

There is further evidence for fish withdrawing from noxious events. In a study by Chervova and Lapshin (2011), common carp withdrew from electrical stimulation, with reduced avoidance responses after anaesthetic was administered (while normal motor activity was unaffected). In another study, goldfish learned to avoid electric shocks (Yoshida and Hirano, 2010). These two studies provide examples of teleost fish finding a noxious stimulus so aversive that they altered their behaviours to avoid it. In addition, the avoidance response to a novel object was seen to be impaired in rainbow trout treated with a noxious stimulus (Sneddon, Braithwaite, & Gentle, 2003b). To determine whether this impairment was due to the fish being ‘distracted’ by the experience of pain, this impairment would be able to be reversed with the administration of some form of pain relief. The study found that providing analgesic decreased the impairment of the avoidance response, resulting in a return of the avoidance behaviour to the novel object. Changes in behaviours that occur in fish after potentially painful treatments, which are then reduced by painkillers, strongly suggest that such behaviours are a direct result of the painful experience (Mettam et al., 2011; Sneddon, 2003). The studies presented above provide solid arguments that fish are capable of pain perception.

1.4.4 Arguments against pain perception in fish

Certain views, however, criticise these approaches to demonstrating pain perception in fish, and instead propose comparative neuroanatomical studies for forming their basis behind why pain perception is not possible within fish (Rose, 2002; Rose *et al.*, 2014). Rose argues that ‘implicit’ learning is a virtually universal ability of vertebrate and invertebrate animals, and that this type of learning does not demonstrate a capacity for consciousness (Rose, 2002). Rose also provides examples in which unconscious humans, due to massive damage in the cerebral cortex, can still show facial/vocal/limb responses to nociceptive stimuli (Rose, 2002), as well as associative learning or Pavlovian/instrumental types of learning being within the capacity of decorticate, decerebrate, and spinally transected animals (Rose *et al.*, 2014). The position here is that behavioural responses to noxious stimuli do not by themselves necessitate the existence of conscious awareness of pain or feelings (Rose, 2002; Rose *et al.*, 2014).

Brown (2015) and Sneddon (2015) argue otherwise, stating that it would be unreasonable to separate the physical detection of pain (nociception) from the emotional or cognitive responses as they are part of an integrated motivational system which has evolved to reduce chances of injury. As Dawkins (2001) suggests, consciousness is a Darwinian adaptation which has evolved by natural selection. Even the simplest emotional responses to an affective state (e.g., pain or fear) are widespread amongst vertebrates, which is to be expected considering the great fitness value that comes with such an integrated system (which is to protect the animal from future harm).

Rose (2002) also affirms that human capacities of language and consciousness have resulted from the later, very separate evolutionary development of a complex and enlarged brain. The cerebral hemisphere size and complexity of the mammalian brain is presented here as the principle difference from that of other vertebrates, with the development of the neocortex being the distinguishing feature. Rose advocates that the existence of consciousness requires widely distributed brain activity that is complex and temporally coordinated, and thus requires extensive neurological ‘hardware’ and ‘software’ to accomplish this function (Rose, 2002). Evidence is presented to demonstrate that the capacity for human conscious awareness is dependent on the human neocortex, satisfying such functional criteria due to its unique structural features. It is then maintained that, since the brains of fish lack the complexity in the structural features required for the generation of consciousness as we understand it, they are therefore incapable of perceiving pain (Rose, 2002).

Some argue that such a comparative analysis between fish and human is inappropriate (Braithwaite and Huntingford, 2004; Brown, 2015). This “Hardware-dependent explanation” (Dawkins, 1998, p.324) taken by Rose assumes that conscious experiences are unique to specific brain structures (the neocortex in this case) and ignores the possibility that, in species that are very different from us, other brain pathways could give rise to consciousness (Dawkins, 1998; Brown, 2015).

Such an anthropocentric approach is flawed for the following reasons. Instead of following a linear progression from inferior to superior vertebrates with humans at the current peak, the evolution of vertebrates is random with highly diverse groups each having their own specialisations (Brown, 2015). Each species is specifically tuned to match the niche it occupies. The biological complexity of the animal should therefore not be defined by how closely related the animals are to humans, but by the niche they occupy and the problems they face during their daily existence (Brown, 2015).

Many scientists believe that there are multiple levels of consciousness (Primary, Secondary, and Tertiary), and the sorts of processing associated with fear and pain are almost definitely associated with the primary-process consciousness that are likely widespread amongst vertebrates (Panksepp, 2005). It is perfectly reasonable to conclude from Rose's rationale that fish are not capable of perceiving pain in the same capacity of conscious awareness as humans are. However, to make a definitive conclusion on a fish's general capability to perceive pain at any level from a comparative analogy to humans is flawed for the simple reason that the two are too different from each other in terms of evolutionary life history. Rose makes the important point himself, stating that to the extent that human and fish brains differ, the properties of putative consciousness in humans and fishes will differ as well (Rose *et al.*, 2014). While it is likely that fish experience consciousness in a different manner to that of humans, it is arguably a leaping assumption to then claim that they are incapable of perceiving any kind of pain.

1.4.5 Additional evidence and final points

Ultimately, all of the measures currently proposed for identifying consciousness in fish have yet to directly accomplish this, as posited by Rose (2002) and Dawkins (1998). Dawkins (1998) made the argument that, although rats and humans have striking similarities in response to situations like hunger and cold, it does not follow that they are both consciously experiencing hunger and cold. There is still the possibility that these measures (e.g. avoidance behaviours) could be occurring in animals which simply use the same neural pathways that humans use for automatic unconscious actions, programmed to respond adaptively for the simple benefit of the animal's fitness (Dawkins, 1998). Either conscious experiences are present in some level in many vertebrates, or basic physiological and behavioural mechanisms existed long before consciousness evolved, and all that consciousness did was enhance the ability to deal with longer time scales or greater complexity (Dawkins, 1998).

A recent study by Corder et al. (2019), however, has provided a way of gaining empirical evidence that conscious experiences are present in a vertebrate. This study identified a distinct neural ensemble in the basolateral amygdala of rats that encodes the negative affective valence of pain. These results have begun to refine our neurophysiological understanding behind the multiple dimensions of pain and reaffirm that the measures used to examine affective states and behavioural changes in rats (some through analogy from human behaviour) were appropriate. If such work is replicated in the brains of other vertebrates, such as fish, this could provide an important stepping-stone in proving their sentience. However, the fact that we have not yet been able to definitively prove that fish are sentient does not mean that the consideration of conscious experiences should be ruled out of the science of fish welfare, especially when the arguments provided here strongly point towards that fish are capable of perceiving unpleasant states to at least some extent.

1.5 Importance of welfare in fish farming – salmon as a case study

1.5.1 Animals in a rapidly growing industry

Aquaculture has played a vital role in global food security for decades (Tidwell and Allan, 2001). With increasing demand for affordable, healthy food, the contribution of aquaculture to the total global production of aquatic animals (capture fisheries and aquaculture combined) has risen from 26% in 2000 to 59% in 2020, increasing to 88 million tonnes (FAO, 2022). The growth of the Scottish salmon industry has reflected this development through increased intensification of production practices. Since 1990, there has been at least a six-fold increase in the number of juvenile fish produced per m^3 of water (Ellis *et al.*, 2016). In 2021, production of Scottish Atlantic salmon reached an all-time high of 205,393 tonnes, with more than 50 million smolts transferred to sea in the same year (Munro, 2022). This total tonnage of Scottish farmed salmon, relative to the number of employees on-site, has increased 11-fold within seawater and 6-fold within freshwater between 1985-2016 (Ellis *et al.*, 2016).

The increasing reliance on aquaculture as a food source, and the resultant intensification of the industry to meet these demands threatens the ability of farms to maintain appropriate conditions for fish. The ethical implications alone in dealing with millions of animals on a yearly basis provides a compelling argument for the importance of farmed salmon welfare. Accordingly, fish welfare has gained increasing attention alongside this rapid growth (Ashley, 2007). Salmon welfare is now a significant factor in the industry’s success, affecting public perception and consequently product acceptance and marketing (Broom, 1999; European Commission, 2016). Furthermore, production efficiency, product quality and quantity are also directly related to welfare standards (Southgate and Wall, 2001; FSBI, 2002). In addition to the ethical and financial incentives behind safeguarding salmon welfare, the welfare of any fish is also governed by a legal framework within the UK.

1.5.2 Welfare legislation

On the international level, farmed fish have received little to no serious legal protection or consideration during husbandry or on-farm practices (Giménez-Candela, Saraiva and Bauer, 2020). It was only in 2008 that the World Organisation for Animal Health (OIE) adopted standards on the welfare of farmed fishes in the OIE Aquatic Animal Health code. As no enforcement body was involved in this, however, the OIE's codes and standards are still recommendations with no legal binding weight (Giménez-Candela, Saraiva and Bauer, 2020).

The first main pieces of legislation introduced to cover UK farmed animal welfare were the Protection of Animals Act 1911 and the Protection of Animals (Scotland) Act 1912 (Voas, 2008). Considering the recent establishment of fish farming at the time, this legislation was not intended to cover farmed fish. However, under the latest legislation, the Animal Welfare Act (2006) and the Animal Health and Welfare (Scotland) Act (2006), farmed fish are covered as 'vertebrates'. This offers fish basic protection against unnecessary suffering and places a duty on those responsible to ensure the needs of fish are met.

In comparison, the first legislative protection for farmed animal welfare in the EU began in 1974 with the Protection of Animals in Slaughterhouses Act (UK Parliament, 1974), and it was only until 2009 that fish were recognised as sentient beings under the original Article 13 of the Treaty on the Functioning of the European Union (TFEU) (Council of the European Union, 1998). Still in the UK today, the definition of 'farmed animal' under the Welfare of Farm Animals (England) Regulations (2007) (and similar legislation in Scotland and Wales) explicitly excludes fish. Similarly, the general principles laid down in EU legislation also leave room for interpretation, or are not applicable to fish welfare. The Council Directive 98/58/EC concerning the protection of animals kept for farming purposes lays down the minimum standards under which farmed animals (including fish) are allowed to be bred and kept on farms. However, only specific articles must be applied to fish, with Article 4 (in which further requirements are laid down on farming conditions) explicitly excluding fish (Council of the European Union, 1998). Fish are, therefore, not yet offered the more detailed welfare protection provided to most terrestrial farm animals.

In addition to general aspects of farming in the UK, the Welfare of Animals (Transport) (Scotland) Regulations 2006 (and equivalent legislation in England and Wales) applies to fish, requiring drivers of vertebrate animals to be trained and certified appropriately to prevent unnecessary suffering during transportation. The Welfare of Animals (Slaughter or

Killing) Regulations (1995) requires sufficient training for those carrying out slaughter methods to again prevent any unnecessary suffering. Provisions for controlling fish health and disease are covered in the Aquatic Animal Health (Scotland) Regulations 2009 (and similar legislation in England and Wales). The current Animal Welfare Acts regarding husbandry, transport and slaughter of fish place a duty on those responsible for the fish to ensure their needs are met. These needs, set out in legislation, have been particularly influenced by the principles of the ‘Five Freedoms’, outlined in the Brambell Committee Report (Brambell 1965, cited in Voas 2008). These five freedoms are:

1) Freedom from thirst, hunger, and malnutrition.
2) Freedom from discomfort due to environment.
3) Freedom from pain, injury and disease.
4) Freedom to express normal behaviour for the species.
5) Freedom from fear and distress.

These freedoms, although basic, are based on fundamental, timeless principles (Webster, 2016). However, the Five Freedoms alone are not sufficient to ensure proper animal welfare. For example, the temporary elimination of such unavoidable parts of life (such as fear, pain, thirst, hunger, discomfort etc.) does not by itself generate positive experiences for the animal (Mellor, 2016). This is reflected in the legislation; although the Animal Welfare Act includes a vague “duty of care” to promote going beyond simply avoiding negative conditions, there is currently no explicit requirement to promote positive animal welfare (e.g., through enrichment). One could argue that current legislation only requires fish welfare to, at best, be neutral. There is also no current legislation that dictates any specific conditions under which fish should be kept (FAWC, 2014).

Attempts to strengthen the principles of the five freedoms by adding this detail can often have the opposite effect: the more one expands the argument by adding examples, the more likely one is to leave things out (Webster, 2016). In the context of welfare legislation, however, the Five Freedoms could provide the ethical guidelines from which the legislation of species-specific needs could be built upon. In addition to explicit requirements for positive welfare, this could include the general requirements of the sort included in the Welfare of Farmed Animals Regulations (of which fish are currently excluded): staff competence, record keeping, maintenance and testing of equipment could also be extended to farmed fish (FAWC, 2014). The numerous incentives to improve farmed salmon welfare, along with regulating bodies wishing to avoid additional legislative control, has encouraged the Scottish salmon industry to adopt various standards that promote salmon welfare beyond what is required by the current legislation (FAWC, 2014).

1.5.3 Welfare standards – certification schemes and codes of good practice

Several salmon farming standards currently exist, each with their own set of requisite criteria permitting certification. Criteria for these standards varies, with different emphasis on sustainability, product quality, and welfare. Certifying around 70% of Scottish salmon farms, the ‘RSPCA Welfare Standards for Farmed Atlantic Salmon’ are specifically focused on salmon welfare assurance (Rey Planellas, Little and Ellis, 2019; Salmon Scotland, 2020a; RSPCA, 2021). This certification scheme provides detailed species-specific requirements for health management, husbandry practices, equipment, feeding, environmental quality, vaccination, transport, slaughter and harvest, and handling. RSPCA welfare standards are based on scientific, veterinary, and practical expertise, utilising numerous animal based welfare indicators along with indirect, environmental welfare indicators (Noble *et al.*, 2018).

Another salmon welfare relevant standard is the GLOBAL G.A.P. Aquaculture standard, which is part of an integrated assurance scheme that can be applied to any farm system; this specific ‘module’ provides an extensive checklist for measures which maintain fish welfare along all points of the production chain (GlobalG.A.P., 2017). This includes monitoring stock origin, health management, feeding, welfare risk assessments and numerous other procedures on the farm. The focus of this standard, however, is largely on staff training, record keeping, and maintaining equipment and farming routines (primarily a list of environmental or resource based indicators) and consequently does not comprehensively cover how to assure animal welfare (Noble *et al.*, 2018). The Code of Good Practice for Scottish Finfish Aquaculture (Scottish Salmon Producers Organisation, 2015), while similar to the GLOBALG.A.P. standard, includes many more production stage-specific requirements for fish welfare. This includes water quality, monitoring recommendations and biosecurity. The top six producers of salmon in Scotland (Scottish Salmon Company, Scottish Sea Farms, Mowi, Cooke Aquaculture, Loch Duart, Grieg Seafood), all of which are members of Salmon Scotland, subscribe to this Code of Good Practice.

The Best Aquaculture Practices (BAP) Certification Standards and Guidelines for Salmon Farms is an international certification scheme developed by the Global Aquaculture Alliance (GAA) (Global Aquaculture Alliance, 2016). While the majority of this standard emphasises environmental standards, an ‘Animal Health and Welfare’ chapter is included. Its requirements include a brief list of behavioural indicators, adequate facilities and water quality management, morphological deformities, and handling procedures. Although primarily focused on environmental impacts from aquaculture, the ASC Salmon Standard also has certain criteria relevant to fish welfare, requiring regular veterinarian visits, health and water quality management plans, disease monitoring, and mortality limits (ASC, 2019).

All of these standards recognise that certain factors, such as water quality, are key to fish health and by effect to fish welfare. Accordingly, they contain general requirements to monitor and maintain such factors. How these standards focus on welfare, however, varies significantly (FAWC, 2014). Due to the voluntary nature of these welfare standards, salmon farming companies are free to obtain whichever specific accreditation they deem most appropriate for them. This has allowed for inconsistencies between certification schemes, with no clear benchmark for welfare standards currently existing. The FAWC (2014) states the importance of having welfare related labelling clearly reflect what welfare standards have been achieved. This way, consumers can identify such standards and comparisons between products can be made, and such uniformity would inevitably drive up standards in the whole industry (FAWC, 2014).

1.6 Factors influencing welfare

There are a number of key welfare concerns that are associated with practices that are central to fish farming (FAWC, 2014; Noble *et al.*, 2018). To ensure a comprehensive appraisal of farmed salmon welfare, it is important to cover the entire production process so that any factors which may influence their welfare are considered. Bergqvist and Gunnarsson (2013) describe how threats to salmon welfare in the production process can be divided into four stages: breeding, growth period, capturing alongside transportation, and slaughter. First, eggs and sperm are extracted from anaesthetised fish, followed by incubation in oxygenated freshwater, hatching, and then rearing in flowing water (Santurtun, Broom and Phillips, 2018). Fingerlings, known as parr, are transferred to larger freshwater tanks or cages, where they remain until smoltification (a physiological adaptation from freshwater to seawater). These smolts are then transported to large, floating cages in sheltered bays or sea lochs (sea cages, which are less sheltered), where they grow for one to two years before slaughter. Alternatively, a small proportion of salmon are grown in large enclosed tank systems throughout their entire life (Santurtun, Broom and Phillips, 2018).

Noble et al. (2018) distinguishes welfare concerns (or 'needs') between those that are 'ultimate', which are immediately essential for welfare and survival (e.g. respiration, thermoregulation, body integrity, nutrition), or 'proximate', which improve the ability for long term success (e.g. behaviours that improve body control or strength, exploratory behaviours that improve chances of finding food). While some of these concerns or needs are critical for the salmon at all life stages (e.g. respiration), the importance of some behavioural needs may depend on one or more life stages (e.g. sexual behaviour), or as a form of preparation for a later life stage (e.g. salmon jumping behaviour) (Noble *et al.*, 2018). This means that, while certain factors influence welfare on a continuous basis and must always be monitored, others may not be crucial at every moment.

1.6.1 Breeding and genetics

Genetic aspects (e.g. variation) and reproductive practices (e.g. handling, environmental effects during hatching, larvae feeding) are critical to fish welfare (Bergqvist and Gunnarsson, 2013). Selective breeding has shown to have positive welfare effects, producing less aggressive and less excitable Atlantic salmon which would be better suited to artificial rearing conditions and handling (Håstein, Scarfe and Lund, 2005). Selection has also produced disease-resistant fish (e.g. for vibriosis, furunculosis) (Håstein, Scarfe and Lund, 2005; Brown *et al.*, 2008). However, the effects of genetic manipulation on fish welfare depend on which genes are modified, and there are concerns about unforeseen phenotypic consequences such as deformities which could impact feeding or respiration (Håstein, Scarfe and Lund, 2005). Numerous malformations of the spine, common in farmed fish, are considered in part to be effects of hereditary factors (e.g. inbreeding) (Bergqvist and Gunnarsson, 2013). Intensive manual handling of broodfish can also be a stress to the fish, and anaesthetics prior to handling to account for this must also be considered in the interest of their welfare (Cooke, 2017).

1.6.2 Growth period

For most fish in aquaculture, their growth period represents the longest stage in the life of the fish (Bergqvist and Gunnarsson, 2013), with farmed Atlantic salmon being grown for as long as three years (longer than many terrestrial farm animals) (FAWC, 2014). This stage in the production process accordingly requires significant consideration, as chronic welfare issues can have a much larger welfare impact (FAWC, 2014). Intensive aquaculture production can threaten fish welfare by subjecting fish to certain stressful environmental (e.g. water quality) and health conditions (e.g. physical injury, infectious diseases) (Oliva-Teles, 2012). Such conditions compromise the functional and affective state of the fish. In addition, fish behaviour is also closely related to the production process, with numerous implications for welfare (Conte, 2004; FAWC, 2014). Although these various factors that affect salmon welfare have been divided into different stages, they interact with each other (Ashley, 2007; Santurtun, Broom and Phillips, 2018) and should therefore be considered together rather than individually.

1.6.2.1 Health and nutritional factors

Poor health causes immediate impacts to welfare due to the diminished functional state of the animal. Physiological stress can potentially lead to further hazards to welfare through a number of mechanisms. This includes impaired responses to further stress, negative social interactions, reduced feeding, reductions in immunocompetence, and consequently increased susceptibility to pathogens, disease and further suffering (Ashley, 2007). Good welfare thus involves minimising and preventing the occurrence of such stressors (e.g. injury and disease, all of which have a potential to occur within many aquaculture practices (Ashley, 2007).

Disease: Farmed salmon are vulnerable to a variety of harmful infectious diseases (e.g., parasites, bacteria, fungi and viruses) and non-infectious diseases (e.g., disorders related to poor husbandry conditions). Each disease is capable of causing signs that are clear indicators of poor welfare and potential suffering (Bergqvist and Gunnarsson, 2013; Cooke, 2017; Noble *et al*., 2018). The impact of a disease on the health and welfare of salmon in a cage will depend on the type, intensity, and duration of the disease, along with the proportion of fish affected (Nob*le et al*., 2018).

Non-infectious diseases (environmental, nutritional, and hereditary): Diseases associated with production practices in Atlantic salmon are often a product of poorly managed nutritional, environmental, or hereditary factors (Ashley, 2007). Nutritional diseases arise from toxins, deficiencies of micronutrients, or even malnutrition (lacking or excessive). These diseases include

1) Cataracts due to deficiencies in histidine (Ersdal, Midtlyng and Jarp, 2001; Santurtun, Broom and Phillips, 2018)
2) Numerous factors implicated with spinal deformities, such as phosphorous deficiencies leading to scoliosis (Silverstone and Hammell, 2002; Håstein, Scarfe and Lund, 2005)
3) Rancid feed can cause fatty liver syndrome (Håstein, Scarfe and Lund, 2005)
4) High-energy diets have also been linked to fatty deposits in the cardiac ventricles of farmed salmon, potentially predisposing them to cardiac disease (Santurtun, Broom and Phillips, 2018)

Opercular deformities have been associated with inappropriate ambient temperatures while rearing fry (Poppe, Barnes and Midtlyng, 2002), while lower jaw deformities have been linked to the use of triploids (Amoroso *et al.*, 2016). Other soft tissue malformations, including eye lesions, swim bladder deformities, and heart deformities (e.g. hypoplasia or *situs inversus)* have also been reported in farmed Atlantic salmon (Håstein, Scarfe and Lund, 2005; Ashley, 2007).

Apart from likely causing some level of suffering, diseases that impair the function of the salmon, such as cardiac abnormalities, often lead to reductions in size and stress tolerance. As a result, these salmon are typically the first to die during stress related practices (Håstein, Scarfe and Lund, 2005; Ashley, 2007). Fin damage is another common ailment with farmed Atlantic salmon that involves various lesions to the fin, and is usually a result of abrasion with the environment or aggressive interactions (Turnbull, Richards and Robertson, 1996; Ashley, 2007). Like many other non-infectious diseases, fin damage can increase susceptibility to infectious diseases by other pathogens and parasites, further compromising salmon welfare (Turnbull, Richards and Robertson, 1996; Ashley, 2007).

Infectious diseases (bacteria and viruses): Farming fish in dense populations within exposed environments inevitably leads to outbreaks of infectious diseases (Robertsen, 2011). The use of vaccines has helped prevent bacterial diseases, such as vibriosis and furunculosis, from devastating Atlantic salmon farming (Robertsen, 2011; Noble *et al*., 2018). Viral diseases, however, present a much larger threat to the health and welfare of farmed Atlantic salmon (Ashley, 2007; Noble *et al*., 2018). This is largely due to the lack of effective vaccines that are available (Noble *et al.*, 2018). Viral diseases (and the involved viruses) that are of particular importance in salmonids include (Ashley, 2007; Robertsen, 2011; Noble *et al.*, 2018; RSPCA, 2018a)

1) Infectious pancreatic necrosis (IPNV)
2) Infectious salmon anaemia (ISAV)
3) Heart & skeletal muscle inflammation - HSMI (Piscine orthoreo virus)
4) Pancreas disease (Salmonid alphavirus / Salmon pancreas disease virus)
5) Cardiomyopathy syndrome – CMS (Piscine myocarditis virus)
6) Salmon gill poxvirus (SGPV)
7) Viral haemorrhagic septicaemia – VHS
8) Infectious haematopoietic necrosis (IHNV)
9) Sleeping disease (SAV)

Although salmon appear to have a strong innate immunity against viruses, owing to their well-developed interferon system, these viruses have caused high mortalities in salmon farming (Robertsen, 2011). This suggests that one of two situations which may be occurring: Viral outbreaks may be due to the introduction of new pathogens, the mutation of existing pathogens, or new routes of contact between hosts. Alternatively, the conditions presented by certain production systems may be resulting in changes (suppression) in immunity. Avoiding situations which lead to the suppression of the immune system, in order to help prevent the prevalence of such infectious diseases, is therefore a priority in safeguarding salmon health and welfare.

Infectious diseases (parasite and fungi): Amoebic gill disease (AGD) is another serious disease issue threatening farmed Atlantic salmon, caused by the amoeba *Neoparamoeba* *perurans* infecting the gills (Powell, Reynolds and Kristensen, 2015; Noble *et al*., 2018). This disease causes massive inflammation of the gills, affecting respiration, reducing appetite, and leading to severe mortality rates if left untreated (Powell, Reynolds and Kristensen, 2015; Noble *et al.*, 2018).

Another widespread threat to the health and welfare of salmon are sea lice *Lepeoptheirus salmonis* (Ashley, 2007; Stien *et al.*, 2013; Powell, Reynolds and Kristensen, 2015; Cooke, 2017). At the initial infective copepod stage, where feeding on the salmon has yet to occur, salmon already exhibit a primary stress response, evident by elevated blood cortisol and glucose (Stien *et al.*, 2013). Once sea lice develop to the feeding stage, these parasites cause damage to the fish's skin, scales, and mucous. The loss of such physical and chemical barriers to the environment compromises osmoregulation and can act as a vector of disease to the fish. This, in addition to inflammatory responses, changes in appetite, osmotic disturbances, and delayed healing of injuries means that sea lice can severely impact fish health and welfare and eventually lead to mortalities (Håstein, Scarfe and Lund, 2005; Ashley, 2007; Stien *et al.*, 2013).

Infection with the *Saprolegnia* oomycete can cause serious disease conditions (e.g. development of serious skin, fin, and tail lesions) in the freshwater stages of salmon production, being particularly severe at times when the fish's resistance to infection is compromised (Cooke, 2017; Noble *et al*., 2018; RSPCA, 2018).

Disease treatment and vaccination: Although the prevention and treatment of disease is an integral part of safeguarding salmon welfare, their welfare must also be monitored and protected during these practices as they can also be stressful to the fish (Huntingford *et al.*, 2006). Delousing treatments, bath treatments, and vaccinations can involve handling out of water, the use of anaesthesia, and injections, all of which can cause severe stress to the fish if done poorly (Ashley, 2007; Berg, Haagensen and Horsberg, 2012; Stien *et al.*, 2013).

Feeding practices: Successful feeding is rewarded by replacing the feeling of hunger with satiation, and fish have shown strong anticipatory behaviour for preferred food sources, indicating an emotional qualitative component of wanting/liking (Warburton, 2003). Inappropriate diets also increase disease susceptibility and negatively alter the behaviour of fish (Oliva-Teles, 2012; Cooke, 2017; Sloman *et al.*, 2019). Adequate nutrition for Atlantic salmon at their species and life stage–specific needs is therefore a key component in protecting their welfare (Oliva-Teles, 2012; Noble *et al*., 2018).

1.6.2.2 Environmental factors

Water quality: The physico-chemical characteristics of water (i.e. 'water quality') have a profound impact on the biological functioning in Atlantic salmon (Brown *et al.*, 2008), and they are accepted as one of the most significant environmental factors for salmon welfare (Huntingford *et al*., 2006; Bergqvist and Gunnarsson, 2013; FAWC, 2014; Cooke, 2017; Noble *et al*., 2018; Santurtun, Broom and Phillips, 2018; Sloman *et al*., 2019). Safeguarding the health and welfare of salmon through water quality requires appropriate levels of O_2, metabolic wastes (CO_2, Ammonia/Nitrite), salinity, toxins, temperature, and pH (Conte, 2004; Huntingford *et al*., 2006; FAWC, 2014; Noble *et al*., 2018). Many of these parameters interact with each other, with optimal ranges depending on numerous factors. For example, CO_2 levels and its effects are affected by pH, temperature, hardness of water, water flow and stocking density (Brown *et al.*, 2008; FAWC, 2014). The available dissolved oxygen in water depends largely on temperature, salinity, aeration, and partial pressure of oxygen in the air in contact with the water (Brown *et al.*, 2008; FAWC, 2014). Biofouling (algae accumulation on nets) can affect movement of free water (Brow*n et al*., 2008). Poor water flow can also cause localised O_2 depletion and CO_2 accumulation in sea cages, while algal blooms can affect pH balance and collapsed blooms can deplete O_2 levels and release ammonia (Cooke, 2017). With insufficient oxygen levels, hypoxia can cause a stress response in salmonids (McNeill and Perry, 2006; Remen, 2012), and salmon can die within minutes of not respiring (Stien *et al.*, 2013). Respiration can also be limited during handling or from non-functional gills as a result of injury, disease, or parasites (Noble *et al.*, 2018).

Imbalances of these parameters can cause direct harm to the fish through disruption of physiological functions, such as ionic regulation, gill and kidney function, or by destroying the fish's mucous coating (Conte, 2004). Poor water quality can also affect a salmon's immunocompetence, growth, and survival (Santurtun, Broom and Phillips, 2018).

Temperature is another important environmental factor influencing salmon biology (Stien *et al*., 2013; FAWC, 2014; Noble *et al*., 2018a). Being poikilothermic, the body temperature of salmon is regulated by ambient water temperature, which can therefore only be controlled by swimming to other available areas with the most appropriate temperature (Noble *et al.*, 2018). Temperature, together with oxygen, determines the metabolic rate of salmon and acts as a controlling factor for the salmon's physiological performance, including their capacity for dealing with other stressors (Stien *et al.*, 2013).

Temperature and light have other direct implications for salmon behaviour and welfare. Oppedal et al. (2007) found a behavioural trade-off in Atlantic salmon between preferences for temperatures in a thermally stratified environment and attraction to brighter parts of the cage. Crowding due to competition for favourable conditions (e.g. appropriate temperature and light levels) could likely be a more serious welfare concern than stocking density in and of itself, and sites with strong vertical temperature stratifications should take the possibility of such schooling densities occurring into account (Oppedal, Juell and Johansson, 2007). When salmon transition from freshwater to seawater, appropriate photoperiods, temperature, and salinity conditions are required for proper timing and completion of events such as smoltification and sexual maturation (Brown *et al.*, 2008).

Being anadromous, Atlantic salmon are also under threat from osmotic stress, particularly during the transfer of smolts to sea. Fish that are not physiologically ready to move to seawater can suffer from hyperosmotic stress, often dying as a result (FAWC, 2014; Noble *et al*., 2018). Conversely, there is the danger of smolts reverting back to freshwater physiology if kept in freshwater for too long (Stien *et al.*, 2013; FAWC, 2014; Noble *et al.*, 2018). Smoltification, however, is becoming a more effectively managed process through which the industry is using environmental and dietary manipulation in an attempt to increase uniformity of fish before transfer to seawater (FAWC, 2014).

Stocking density: The deterioration of water quality is also directly proportional to the biomass and metabolism of the salmon in relation to the volume and turnover of water (Håstein, Scarfe and Lund, 2005). The impact that the stocking density has on fish welfare is difficult to assess, due its complex nature and the numerous interrelated factors involved (Håstein, Scarfe and Lund, 2005; Bergqvist and Gunnarsson, 2013). However, stocking density is closely related to water quality (Ashley, 2007; FAWC, 2014) and affects other aspects of fish welfare at all life-cycle stages (Håstein, Scarfe and Lund, 2005; Bergqvist and Gunnarsson, 2013; FAWC, 2014; Cooke, 2017). In addition to deteriorating water quality, inappropriate stocking densities can severely impact the welfare by increasing agonistic behaviours between individuals, leading to poor body condition and increased stress levels (Turnbull *et al.*, 2005; Cooke, 2017; Santurtun, Broom and Phillips, 2018).

Reduced access to food is another possible consequence of inappropriate stocking density which, in combination with open wounds and increased stress levels, can lead to increased susceptibility to disease (Cooke, 2017). The FAWC (2014) also suggest that sufficient space is required to permit normal behaviour and minimise pain, stress and fear of the fish, although this will depend on a number of conditions. While Turnbull et al. (2005) found that densities above 22 kg/m^3 impaired the welfare of Atlantic salmon, the authors determined that the various factors connected to density means that this value may be appropriate for some farms but not others depending on different farm practices and conditions.

Predators: Aside from injuring or killing salmon in sea pens, the presence of predators can also have major welfare impacts by causing fear and stress (FAWC, 2014; Cooke, 2017). This is often manifested by behavioural changes and/or reduction in feeding (Cooke, 2017).

Environmental enrichment: In addition to stocking density, Sloman et al. (2019) asserts the importance of environmental enrichment for the welfare of fish under our care. The natural environments of fish often have spatial and temporal variations in variables such as temperature, light levels, and current speeds (Oppedal, Dempster and Stien, 2011), while the environments that farmed fish tend to experience (e.g. tanks and sea cages) are simple in design and relatively uniform in comparison (Huntingford *et al*., 2006; Noble *et al*., 2018). Gradients are important for fish to optimize certain factors (temperature, current velocity etc.) and acquire certain information (regarding hazards, feed acquisition etc.) (Noble *et al*., 2018). In order to permit fishes to perform more natural behaviours, then how they are housed must be taken into consideration (Sloman *et al.*, 2019). This raises questions whether such environments allow sufficient variation for the needs and preferences of the captive fish (FAWC, 2014). Fish in their natural habitats exhibit preferences for specific environments, and there are reports linking improved culture performance from accommodating for fish’s behaviours (Conte, 2004). Environmental and feeding enrichment strategies used during rearing of Atlantic salmon appear to also improve their survival rates, and may help to reduce any deleterious behaviour (Brown, Davidson and Laland, 2003; Ashley, 2007). A study from Näslund et al. (2013) suggests that enrichment may also improve salmon welfare by helping to reduce the impact of stressors experienced in hatcheries. Tank design, water flow, and the availability of shelters are some of what need to be considered relative to the species (Huntingford *et al.*, 2006; Sloman *et al.*, 2019).

1.6.3 Behaviour

Many stressors are intimately linked with the behaviour of salmon; while stress can initiate behavioural changes, forced behavioural changes can also cause stress of their own to the fish (Conte, 2004). Managing this species-specific behaviour, either by supporting the behavioural needs of salmon or by preventing deleterious behaviours, is therefore critical to ensuring their welfare (Conte, 2004; Bergqvist and Gunnarsson, 2013; Noble *et al*., 2018). Fish behaviours that are known to be affected by stressors include feeding responses, avoidance behaviours, orientation and taxes (movement in response to a stimulus), swimming performance, and aggression (Conte, 2004; Huntingford *et al.*, 2006). There are a number of concerns involving the behaviour of Atlantic salmon which are discussed below:

Aggression and competition: Agonistic behaviour to conspecifics has often been recorded in farmed fish, particularly in a species with hierarchal social orders like Atlantic salmon

(Ashley, 2007; Brown et al., 2008; Bergqvist and Gunnarsson, 2013). As aforementioned, this behaviour is often a result of improper stocking conditions (inappropriate densities / feeding methods or heterogeneity in size of fish), which can directly affect the welfare of lower ranking subordinates by feed deprivation (due to being outcompeted) and injuries from other individuals, leading to poor growth, increased stress and vulnerability to disease (Conte, 2004; Huntingford *et al.*, 2006; Ashley, 2007; Bergqvist and Gunnarsson, 2013). Fin damage in such situations is generally attributed to increased aggression from conspecifics (Håstein, Scarfe and Lund, 2005; Cooke, 2017; Santurtun, Broom and Phillips, 2018); since the fins of salmonids seem to be highly innervated, and may function as mechanosensory organs, biting/damage to fins is likely to be painful (Santurtun, Broom and Phillips, 2018). Agonistic behaviour and outcompeting during feeding times also means that the increased feed intake by dominant individuals, at the expense of subordinates, causes a size divergence within the group of salmon (Santurtun, Broom and Phillips, 2018). This can lead to a positive feedback situation in which size disparity further increases agonistic behaviour, and the welfare of lower ranking individuals is impacted further.

Behaviour control: Salmon must also able to freely control their bodily movements and positioning, including regulation of buoyancy and movements away from stimuli/perceived dangers (Stien *et al*., 2013; Noble *et al*., 2018). When this ability is hindered (e.g. when fish are crowded or handled), there are significant increases in O_2 consumption, catecholamine, cortisol and serotonin levels, and avoidance behaviours which all indicate stress and potential fear (Noble *et al.*, 2018).

Social contact: The social needs for predictable interactions between Atlantic salmon vary through their life stages, being territorial and aggressive during freshwater periods and changing to schooling behaviours at smoltification (Stien *et al*., 2013; Noble *et al.*, 2018). In a study by Fernö and Holm (1986), the frequency of aggression in juvenile Atlantic salmon was found to have a negative correlation with stocking density. The increase in density was suggested to hinder the establishment of territories between the salmon, which the study proposed was the source of their aggressive behaviours.

Rest: Having opportunities to reduce activity levels is important for maintaining normal body functioning in salmonids (Farrell, Johansen and Suarez, 1991; Stien *et al.*, 2013). Salmon post-smolts reared at higher water velocities have exhibited signs of poor welfare which include reduced growth, skin and fin damage, and lower expression of the behavioural repertoire observed in fish at lower velocities (Solstorm *et al.*, 2015, 2016).

Suppression of behaviour (sexual / feeding / migratory / exploratory): Maturing wild Atlantic salmon seem to exhibit an inherent need to migrate to rivers where they can perform sexual behaviours that include courtship, choosing of mates, and spawning (Thorstad *et al.*, 2011). Confinement to a sea cage may cause suffering if salmon migratory behaviour is based on an intrinsic drive which the salmon are then unable to fulfil (Ashley, 2007). In the wild, fry and parr constantly explore their environment, and this exploratory behaviour also enables the fish to learn the location of refuges within their range (Brown *et al.*, 2008).

Salmon are selective feeders with an ability to distinguish between different types of feed (Brown *et al.*, 2008), and this could provide an opportunity for enrichment via feeding methods (e.g. live prey for salmon parr; (Brown, Davidson and Laland, 2003). Aside from aggression, abnormal behaviours (e.g. atypical swimming) often result from the suppression of certain behavioural needs (Ashley, 2007), which is indicative of stress and poor welfare as the animal makes constant unsuccessful attempts to remedy its situation (Bergqvist and Gunnarsson, 2013).

1.6.4 Handling, transport, and slaughter

The time periods involved in handling, transport, and slaughter are relatively brief compared with the growth period. However, these activities can be damaging, stressful, and result in very poor welfare (Chandroo, Duncan and Moccia, 2004; Håstein, Scarfe and Lund, 2005; Huntingford *et al.*, 2006; Bergqvist and Gunnarsson, 2013). These activities therefore require significant consideration. The concerns related to these procedures are closely linked to many of the factors previously mentioned, a number of which (e.g. water quality, injuries) are more likely to result in harsher consequences due to the conditions involved.

Handling and transport:

The handling and transportation of salmon, whether it be from freshwater to seawater, to stunning and killing facilities, or for routine inspection, is an unavoidable part of the farming process (Brow*n et al*., 2008; Bergqvist and Gunnarsson, 2013; Santurtun, Broom and Phillips, 2018), and involves a number of potential stressors which can affect salmon welfare differently.

These stressors include: crowding, handling, pumping, poor water quality, removal from water, exhaustion, injuries, confinement, and spread of disease (Chandroo, Duncan and Moccia, 2004; Håstein, Scarfe and Lund, 2005; Huntingford *et al.*, 2006; Bergqvist and Gunnarsson, 2013; Santurtun, Broom and Phillips, 2018).

Short-term crowding of fish prior to management procedures, such as transport, can be one of the most stressful stages for salmon (Santurtun, Broom and Phillips, 2018). There are potential decreases in O_2 levels and water quality, along with increased chances of injury through abrasion and possible increased stress responses to further stressors (e.g. net capture) (Ashley, 2007). Along with pumping, handling, and grading of salmon, crowding provides opportunities for damage to the epithelial layer (FAWC, 2014; Santurtun, Broom and Phillips, 2018). Such injuries from these practices often lead to an increased risk of infection owing to the loss of the salmon’s physical and chemical barriers (scales, skin and mucous coat) (Conte, 2004; Ashley, 2007). This is coupled with the fact that such physical disturbances often evoke the neuroendocrine stress response, resulting in increased blood cortisol levels which (if kept at high levels over long periods of time) is associated with decreased disease resistance (immunosuppression; Huntingford *et al.*, 2006; Santurtun, Broom and Phillips, 2018). Consequently, the spread of disease often becomes a serious health and welfare concern during these practices. Transporting or handling salmon usually leads to the fish being out of water for short periods, which can also elicit maximal emergency physiological responses in fish (Ashley, 2007). In addition to physiological stressors and the risk of external wounds, excessive weight when handling fish out of the water can lead to further injuries from compression, including spinal damage (Conte, 2004).

During transportation, poor water quality can adversely affect a salmon’s immunocompetence, seawater tolerance, growth, and survival (Santurtun, Broom and Phillips, 2018). Maintaining appropriate O_2 levels, pH, temperature, and salinity are important in preventing physiological stress to the salmon (Huntingford *et al.*, 2006; Cooke, 2017). The build-up of metabolic wastes in confined spaces, such as ammonia and CO_2, are also essential in minimising stress (Ashley, 2007; Santurtun, Broom and Phillips, 2018). Confinement alone could also be a stressor, leading to increased cortisol levels in some fish species (Huntingford *et al.*, 2006). In order to help maintain good water quality by reducing metabolism and evacuating the fish’s gut, food withdrawal prior to transport and disease treatment is a commonly used practice (Conte, 2004; Ashley, 2007). Being ectothermic, short-term feed deprivation is likely to be less detrimental for salmon welfare, although it is still important to appreciate the effects of starvation and malnutrition, which can include changes in metabolic activity and behaviour related to competition (e.g. potential for increased aggression) (Ashley, 2007; Cañon Jones *et al.*, 2010).

Slaughter:

It is well known that poorly managed slaughter can cause severe negative effects on welfare at numerous stages before the actual death of the fish, including pain, fear, stress, starvation and exhaustion (Conte, 2004; Håstein, Scarfe and Lund, 2005; Huntingford *et al.*, 2006; Ashley, 2007; Bergqvist and Gunnarsson, 2013; Cooke, 2017; Santurtun, Broom and Phillips, 2018).

Methods of handling during the transfer to the slaughter facilities up to the point of stunning and loss of consciousness are equally important as the method of slaughter itself, as the handling can cause significant levels of stress (crowding stress, physical injuries, exhaustion all possible if done poorly; Ashley, 2007; Cooke, 2017). Prior to slaughter, Atlantic salmon are often deprived of food or some days to reduce metabolism and evacuate their guts, thus reducing oxygen demand and waste production during transport and handling (Ashley, 2007; Santurtun, Broom and Phillips, 2018). Stunning and loss of consciousness must be confirmed before killing the salmon, and common methods used for Atlantic salmon include automated percussive stunning (with appropriate type of hammer and force used) and electrical stunning with the appropriate electric field (Conte, 2004; Ashley, 2007; Bergqvist and Gunnarsson, 2013; Cooke, 2017; Santurtun, Broom and Phillips, 2018). While these methods are seemingly able to achieve humane slaughter in Atlantic salmon (Conte, 2004; Ashley, 2007), poor stunning can occur (Ashley, 2007; Cooke, 2017) causing the animal to suffer unnecessarily. Loss of consciousness should be confirmed before confirmation of death (Håstein, Scarfe and Lund, 2005; Ashley, 2007), and the technique used to confirm the death of the fish should be done swiftly to avoid regain of consciousness (Ashley, 2007).

1.6.5 Mortality

Most causes of mortality are typically associated with some form of suffering before death (Ellis *et al.*, 2012), and the aforementioned health, environmental, and husbandry factors which could contribute to the death of a salmon are of no exception. Since mortality can be the result of an array of different problems associated with poor welfare, it follows that mortality is another principle welfare issue.

Since high mortality rates can arise during episodes associated with disease outbreaks or poorly managed periods of husbandry, mortality can also serve as an important retrospective welfare performance indicator (Ellis *et al.*, 2012).

1.7 Welfare Assessment

1.7.1 Considerations when evaluating welfare on a farm site

To assess every aspect of salmon welfare, a variety of welfare indicators (WIs) have been established. These WIs are assessments that provide qualitative or quantitative information on different aspects of the animal's welfare, depending on the WI being used. The complex, multi-faceted nature of welfare, combined with the various welfare needs of farmed fish, means that there is no single WI that can cover all the relevant aspects of husbandry systems, farmed species, and situations (Brown *et al.*, 2008; Algers *et al.*, 2009). An accurate assessment of farmed salmon welfare is therefore only possible with a collection of species-specific (and sometimes system-specific) indicators, which would cover the various aspects of welfare previously discussed.

A set of criteria should first be set to determine what indicators to include for evaluating farmed animal welfare (Botreau *et al.*, 2007). This set of criteria should fulfil the following theoretical and practical requirements for on-farm welfare assessment (Botreau *et al.*, 2007; Turnbull and Kadri, 2007; Brown *et al.*, 2008; Algers *et al.*, 2009; Noble *et al.*, 2018):

- Set list of WIs used must be exhaustive; together, they must cover all important aspects of welfare.
- This list must be kept minimal; i.e., containing only necessary, mutually exclusive indicators. No repetitive, redundant, largely overlapping, or irrelevant measures.
- The indicators must be, at least to some degree, independent from one another. I.e., the interpretation of one indicator must not rely on that from another. There should also be as few functional links between welfare indicators as possible.
- WIs must be repeatable on the farm site.
- WIs must be validated, reliable, auditable, and feasible for on-farm use.

From this criteria, the determined set of WIs can be applied to assess the welfare in one of two ways. One option is by how much the fish has deviated from what is the accepted 'norm' for the animal within a 'good' environment. This is not necessarily that which is 'natural' for wild fish: assessments of deviation from normality must rather be based on baseline studies of farmed fish in satisfactory environments (Brown *et al.*, 2008). Alternatively, the determined set of WIs can be assessed by the degree of non-fulfilment of the animal's 'needs' (Morton and Griffiths, 1985; Brown *et al.*, 2008; Algers *et al.*, 2009). When applying these WIs to determine the severity of a welfare problem, it is important to also consider the duration and the number of individuals affected; this is particularly relevant with the large population sizes involved within aquaculture (Turnbull and Kadri, 2007).

Although the consideration and action on fish welfare is often carried out at a group level on fish farms, the consideration of individual fish welfare is equally important, regardless of whether or not they can be monitored to this point (Brown *et al.*, 2008; FAWC, 2014). To help ensure that good welfare is maintained, it may also be important to include indirect WIs that are outside of measuring the fish and their surrounding environment. This could include indicators through 'good practice' (e.g. staff training on welfare, good husbandry protocols, health & contingency plans) (Brown *et al.*, 2008).

1.7.2 Challenges with on-farm assessments of salmon welfare

The FAWC (2014) state how there are a number of fundamental differences between monitoring fish and terrestrial animals. When monitoring fish, a number of these differences have practical problems associated with them, particularly for farmed salmon (some of which technological solutions have been and are being further developed):

- The transition between salt and fresh water has numerous effects that change the relative importance of indicators, such as water quality, delivery of oxygen, and vulnerability to certain diseases and parasites. Ergo, welfare assessments are context dependent
- The 3-dimensional aquatic environment means that salmon are only visible from the surface (also often severely limited) unless aided with monitoring equipment. This adds challenges for the identification and monitoring of individuals, particularly in sea cages with 100,000s of fish. Video filming has been used to reveal the physical status and behaviour of fish, while sonar and echo integration has been used to help visualize the distribution of large numbers of fish (Juell *et al.*, 2003)
- Being stocked at such high numbers and densities also creates practical challenges for how the fish can be properly monitored
- There are a number of behavioural implications for an animal such as salmon (naturally migratory species, poikilothermic animal that controls their physiology by selecting appropriate environmental conditions, hierarchical structure within the species etc.) that must be considered when assessing their welfare

The evaluation of the needs of the salmon, and ultimately its welfare, should also take into consideration how the functioning of the fish (physiological and behavioural) differs drastically during the different life stages of the fish (egg → alevin → fry → parr → smolt → Adult Salmon) (Brown *et al.*, 2008). For example, levels of aggression to conspecifics can be particularly higher in the FW parr stage; schooling behaviours can also depend on group size and life stage, which in turn can also reduce agonistic behaviours in salmon (Brown *et al.*, 2008).

1.7.3 Assessing welfare: welfare indicators

1.7.3.1 Different classes of welfare indicators (OWIs, LABWIs, etc.)

Welfare indicators (WIs) are often classified by how appropriate they are for on-farm use in terms of practicality (Noble *et al.*, 2018). Operational Welfare Indicators (OWIs) are WIs that are feasible enough for staff to use on farm sites (Noble *et al.*, 2018). Laboratory Based

Welfare Indicators (LABWIs), which tend to provide in-depth information to certain parameters of the animal, are WIs that require access to analytical facilities (e.g. laboratory) for their measurement and thus tend to not be appropriate for on-farm use (Noble *et al.*, 2018). OWIs can also be further classified by the degree of labour and time required to carry them out on-site (passive vs. manual OWIs).

The FISHWELL Handbook also presents a simplified scoring system for the ‘operational feasibility’ of WIs, based on sampling and analytical considerations for each WI (Noble *et al.*, 2018). Their scoring system is as follows: 1 = Readily usable on-site; 2 = Usable on site but requires expertise, further data analysis, or specialist equipment; 3 = Can be sampled on-site but must be analysed in a laboratory; 4 = Either unable to sample on-site, or currently requires extended periods of analysis in a laboratory.

Put simply; 1 = Passive OWI; 2 = Manual OWI; 3-4 = LABWI.

WIs can also be classified into different groups by how directly they measure the animal’s welfare. WIs have been broken down into groups below based largely on the FISHWELL Handbook and other literature (Brown *et al.*, 2008; Noble *et al.*, 2018):

1) Animal-based WIs (i.e., direct / outcome-based WIs): Observations made on physiological, morphological or behavioural parameters of the animal

2) Environmental-based WIs (i.e., indirect WIs): Observations made on the surrounding environment

3) Risk-based WIs (i.e., resource / husbandry-based WIs): Observations on the risks posed / minimised on farming processes, farm management, staff training etc. These also act as indirect WIs for the salmon

The following lists of indicators have been extracted from the FISHWELL Handbook and divided into the appropriate groups. Their ‘operational feasibility scores’ have been given corresponding colours to represent which indicators would currently be appropriate as OWIs.

1.7.3.2 Animal-based indicators (direct / outcome “measures”) (Noble et al., 2018)

Table 1-1. Overview of (individual) animal-based welfare indicators, and their corresponding ‘operational feasibility scores’.

Welfare indicator	Score
Sea lice infestation, condition indices (CI, hepato-somatic index, cardio-somatic index), morphological WIs, emaciation state, sexual maturity state, vertebral deformation, fin condition, scale loss / skin condition, snout / jaw condition, opercula condition, eye haemorrhage, handling trauma, feed in intestine, skin colour change.	1
Opercular beat rate, gill bleaching & status, smoltification state, abdominal organs, vaccine-related pathology, blood & muscle glucose / lactate / pH.	2
EEG & ECG, blood cortisol / ionic composition, cardiovascular responses, osmolality, haematocrit.	3-4

Table 1-2. Overview of (group) animal-based welfare indicators, and their corresponding ‘operational feasibility scores’.

Welfare indicator	Score
Mortality rate, surface activity, appetite, scales & blood in water.	1
Behaviour: Abnormal, aggression, emaciated fish, bulk oxygen uptake.	2
Disease / health parameters, slaughter parameters (EEG, ECG, VER)	3-4

1.7.3.3 Risk & environmental-based indicators

Table 1-3. Overview of environmental welfare indicators, and their corresponding ‘operational feasibility scores’.

Welfare indicator	Score
Water quality (temperature, salinity, oxygen, CO_2, pH, alkalinity, turbidity), lighting, stocking density,	1
Total Ammonia Nitrogen (TAN), nitrite/nitrate, water current speed, ammonia	2
Total suspended solids, heavy metals	3-4

Risk-based indicators:

Staff training (feeding routines, handling fish, disease control, interventions etc.), biosecurity measures, maintaining on-farm records and the frequency of handling fish on-site (for grading, treatments, transport etc.) could all be assessed in addition to the previously mentioned OWIs, as these factors ultimately can play a significant role in influencing the state of welfare with these farmed salmon (Brown *et al.*, 2008).

1.7.3.4 Behavioural analyses of farmed Atlantic salmon

There are a number of behaviours that can be evaluated as OWIs, at either the individual or group level, which deserve further elaboration. A salmon’s behaviour will depend on the context, its species-specific behavioural repertoire, and the ability of an individual to adapt at any given moment (Ohl and Van der Staay, 2012). When using behaviour to evaluate the salmon’s ability to cope with its surroundings, it is specifically the change of an animal’s behaviour in response towards certain given stimuli over time (e.g. feeding events) that can inform us about the individuals ability to cope with its surroundings and possible stressors (Ohl and Van der Staay, 2012).

Fish behaviours that are known to be affected by stressors (and thus could potentially be used as OWIs) include various swimming performances, thermoregulation, orientation, avoidance behaviours, feeding responses, and predator evasion (Conte, 2004). The application of Qualitative Behavioural Assessment (QBA) has recently been explored for farmed Atlantic salmon (Jarvis *et al.*, 2021). This technique involves first generating a list of terms to describe the animal's range of behavioural expressions (specifically how it carries out behaviours, rather than what behaviours it carries out), and then scoring each of these terms by the degree to which each are present within the assessment. This first study proved that QBA is applicable for farmed salmon, achieving acceptable inter and intra-observer reliability in QBA scores obtained and significantly correlating these scores with other ethogram-based behavioural measures (Jarvis *et al.*, 2021). However, no study has yet examined QBA's capabilities in capturing changes in emotional expressivity QBA for farmed salmon following exposure to stressors. QBA could prove to be a valuable tool for farmed salmon welfare monitoring, providing a time-efficient, non-intrusive approach to objectively evaluating a range of emotional states that may be present within salmon (Jarvis *et al.*, 2021). Through further validation, the successful inclusion of QBA as such a tool would allow for incorporating a 'feelings-based' approach into farmed salmon welfare assessments. Like any other welfare indicator, however, QBA would need to be used in combination with other welfare indicators to ensure a comprehensive assessment.

1.7.3.5 Implementing OWIs within welfare assessments

As previously mentioned, selecting certain OWIs to use as part of a welfare assessment scheme will likely depend on the production system, the life stage of the salmon, and the specific goals of the welfare assessment (routine monitoring, assessing welfare during intensive practices like treatments or crowding, auditing, etc.; Brown *et al.*, 2008; Noble *et al.*, 2018). Each welfare indicator used will have their own strengths and weaknesses in measuring salmon welfare, along with sampling and analytical considerations that must be acknowledged before choosing to use them (Noble *et al.*, 2018). Furthermore, each welfare indicator will only address certain aspects of welfare and so a variety in OWIs used is vital.

For example, when using morphological OWIs as part of a welfare assessment scheme (e.g. fin damage, skin damage, eye damage, opercular injuries), there are a number of sampling and analytical considerations to take into account (Noble *et al.*, 2018); such OWIs can be qualitatively assessed using observations from above water if the visibility allows for it (or with the use of cameras in real time), and abrupt changes in prevalence can act as an indicator of compromised welfare. However, the severity or frequency of the problem cannot be accurately determined with this method. Alternatively, such OWIs can be quantitatively assessed on a farm site, depending on the sampling and manual handling of the fish at specific times on the site. This is because sampling for this purpose must avoid further harm to the fish. The sample must also be representative of the entire population; this can either be done at opportunistic times (e.g. when all fish are being graded or vaccinated, and there is the possibility of capturing a 'snapshot' of each fish), or the sampling will be time-consuming, labour intensive, and potentially disruptive to existing husbandry tasks like feeding (Noble *et al.*, 2018).

There are obvious strengths to external morphological OWIs. External injuries are immediate indications that welfare has been significantly impacted (Noble *et al.*, 2012). In addition to being easy to observe during routine sampling, increases in injury frequency and severity are quick, feasible, and robust OWIs of poor welfare along with presenting an underlying cause that requires further investigation (Noble *et al.*, 2018). There are, however, also weaknesses associated with such OWIs. Injuries can have various potential causes, and further investigation is required to identify the source of the problem. As previously mentioned, these OWIs can also be very time-consuming, especially in deep sea cages (Noble *et al.*, 2018). Accordingly, the different strengths, weaknesses, sampling and analytical considerations of each OWI that is chosen must all be appreciated within the context of the welfare assessment being carried out.

1.7.3.6 Selecting and combining OWIs based on the purpose of the WAS

As previously mentioned, the purposes behind a welfare assessment scheme can vary greatly, and different purposes may require different approaches behind how salmon welfare is assessed. This can consequently determine the inclusion or exclusion of certain OWIs, and whether they are animal, environmental, or risk-based. Certain approaches may also require a different structure behind how the OWIs are organised and evaluated.

This section will examine this variety, and briefly compare currently existing WASs on how they differ depending on their purposes. This will include:

1) What OWIs are included, and why.
2) The structure of the WAS (i.e., how OWIs are combined into different categories).
3) If there is a scoring system included, how this is implemented.

Example #1 – Salmon Welfare Index Model (SWIM 1.0):

This WAS is designed to enable farmers to conduct a standardised assessment of salmon welfare (specifically in sea cages) using a set list of OWIs (Stien *et al.*, 2013). 'SWIM 1.0' acts mainly as a diagnostic tool, identifying what OWIs are exhibiting reduced welfare on-site. From this, farmers can then address the relevant issues in order to improve the welfare of their salmon. The 17 OWIs included within SWIM 1.0 were selected based on the following criteria:

1) Focusing on ‘quality of life, as perceived by the animals themselves’, certain OWIs would need to cover needs from the animal’s point of view
2) OWIs were to be used specifically by farmers on sea cages
3) OWIs would need to be linked to either:
  - 3a) Physical welfare needs, including respiration, osmotic balance, nutrition, overall health, thermoregulation
  - 3b) Behavioural welfare needs, including control of behaviour, feeding, safety and protection, social contact, exploration, kinesis, rest, sexual behaviour, and body care

The model of assessment is based on the assumption that salmon experience a continuum of welfare states, varying from poor to excellent, and that these states are closely related to the degree of fulfilment of the aforementioned welfare needs. Each OWI in SWIM 1.0 is divided into different levels or ranks, with each rank being assigned an ‘indicator score’. For example, temperature: #1 = 10-15°C, #2 = 7-10°C, #3 = 16-17°C etc. Notably, SWIM 1.0 combines all OWIs (and their corresponding ‘indicator scores’) to calculate a single score (i.e., ‘overall welfare index’) which is then used to represent the welfare status achieved within a sea cage. To determine how much the ‘indicator score’ from each OWI impacts the final ‘overall welfare index’, weighted scores are calculated and used for each OWI. These are based on the supposed intensity, duration, and incidence of the welfare impact that has been linked to each OWI (and its different levels), which was determined through a systematic literature review carried out beforehand (Stien *et al.*, 2013).

There are advantages to providing a single welfare score; standardised scores facilitate comparisons between different sea cages, allows for monitoring changes to overall welfare over time, and makes it easier for stakeholders and consumers to interpret and make comparisons of their own. However, relying solely on a single score may result in the full story of the salmon’s welfare status being poorly represented. In situations where the overall index indicates that an ‘acceptable’ welfare status has been achieved, it is possible that certain OWIs (which have been scored poorly) are masked by other OWIs that have achieved far better scores. In addition, evaluating such scores (and how they are weighted against each other) will inevitably depend on the subjective lines that are drawn between what is “acceptable” or “unacceptable” welfare. A balance should be found in what actionable insights are found from such an assessment, where important aspects of the salmon’s welfare (e.g., behavioural and physical needs) remain separated and the evaluation of each aspect (i.e., whether or not it is acceptable) be context-dependent.

Example #2 – Certification schemes (welfare standards):

Whereas SWIM 1.0 exists as a diagnostic tool, there are other WASs that exist moreso for the purpose of certification schemes, like the ‘Code of Good Practice’ (CoGP) and the RSPCAA welfare standards for farmed Atlantic salmon (Scottish Salmon Producers Organisation, 2015; RSPCA, 2021). In contrast to SWIM 1.0, these certification schemes tend to follow more of a risk-based approach, with the main purposes being to (Stien *et al.*, 2013):

1) Identify hazards, their consequences, and probabilities of occurrence.
2) Find critical control points in the production process.
3) Orient standards around these control points to avoid welfare risks from occurring in the first place (e.g., stress, injury, disease, malnourishment, and mortality).

As the set criteria for these certification schemes prioritise managing the aforementioned control points within the production process, there is largely a focus on farming conditions and husbandry practices that essentially act as indirect OWIs. These criteria involve species-specific requirements, or ‘clauses’, that are organised into different sections within the standards by their relevance to specific husbandry practices or production stages (e.g., sections for farm site locations, stocking density, predator control, handling / crowding / transportation, slaughter, handling mortalities, maintenance of records and equipment etc.) (Scottish Salmon Producers Organisation, 2015; RSPCA, 2021). Farm sites certified under these certification schemes are expected to adhere to the relevant clauses and undergo annual audits to prove their compliance (Scottish Salmon Producers Organisation, 2015; RSPCA, 2021). Instead of calculating an overall welfare index, these audits evaluate farm sites based on their level of compliance to the clauses. Violations, or ‘non-compliances’, range from less severe (i.e., not likely to cause suffering to the salmon, resulting in a formal warning to farm staff) to more severe or repeated non-compliance (i.e., assessors have identified that salmon have likely suffered directly as a result of neglect or malpractice) that may lead to suspension or withdrawal of the certification.

1.7.3.7 Concluding remarks on contrasting WASs

The SWIM 1.0 and certification schemes mentioned are all WASs that recognise certain conditions within salmon farming (e.g., stocking density, stressors, disease, injury, etc.) as key factors that impact salmon welfare. However, the difference in their purposes (i.e., how these WASs approach the monitoring and maintaining of these key factors) results in distinct differences to the selection, structure, and evaluation process of the OWIs involved, even when the same production systems (e.g., sea cages) are involved.

Such contrasts between the different WASs highlights an important consideration when working to develop, evaluate, or improve a specific WAS. Clear guidelines must first be set on what purpose the WAS has; to facilitate routine monitoring on-site, or to act as an auditable set of clauses that provide welfare assurances under a certification scheme?

1.8 The proposed study

With production of Scottish Atlantic salmon reaching an all-time high of 205,393 tonnes in 2021, and over 50 million smolts being transferred to sea in that year (Munro, 2022), concerns for farmed salmon welfare have understandably grown amongst stakeholders and the public (Bergqvist and Gunnarsson, 2013; Barreto *et al.*, 2021). Considering the ample evidence gathered in this chapter suggesting that fish are capable of perceiving their own welfare, there are a number of ethical obligations involved with ensuring the appropriate protection of farmed salmon welfare. In more recent years, these concerns have expanded towards ensuring that welfare assessments for fish also monitor their positive experiences (Fife-Cook and Franks, 2019; Franks, Ewell and Jacquet, 2021; Browning, 2023).

Further developments are therefore required to ensure that the industry is capable of not only monitoring and safeguarding all dimensions of farmed salmon welfare (i.e., both their physical and mental well-being), but are able to do so in a practical, robust manner while providing evidence for this.

However, providing meaningful, industry-relevant contributions towards the monitoring and management of farmed salmon welfare is no simple task. This chapter has already established how complex and multi-faceted the concept of welfare is for any animal, and the anadromous life cycle of Atlantic salmon presents additional complexity to this (Marschall *et al.*, 1998). There are various rearing conditions, husbandry practices, practical considerations, and responsibilities of farm staff that must be considered when monitoring and safeguarding farmed salmon welfare, all of which can be specific to each production stage (Bergqvist and Gunnarsson, 2013; Noble *et al.*, 2018).

In view of this complexity, it was vital that this PhD study utilise both industry expertise and data on current and past welfare practices within the Scottish salmon farming sector to first gain insights into the current state of farmed salmon welfare. Farm staff have extensive hands-on experience in protecting salmon while carrying out husbandry practices (Størkersen *et al.*, 2021). They will thus likely have a unique understanding of the practical limitations involved in implementing welfare monitoring and management practices. In addition, welfare standards are one of the few avenues (outside of legislation) through which assurances can be provided on what level of welfare is actually being achieved for farmed salmon (FAWC, 2014). However, there has been limited research into the underlying frameworks for these standards, what requirements / clauses they consist of (and why), and how farm sites are complying with them.

1.9 Aims and objectives

The overall aim of this PhD study was to provide industry-relevant contributions to the on-farm welfare assessment and safeguarding of Atlantic salmon. An integral aspect of this aim included the validation of a novel welfare assessment tool that would be applicable within an on-farm context.

The following objectives were set out for this thesis:

1) Gather opinions from Scottish salmon farmers in order to:
1a) Evaluate the relative perceived importance that different production stages, husbandry practices, and specific concerns have regarding farmed salmon welfare.
1b) Obtain insights into current welfare practices and challenges associated with monitoring and assessing farmed salmon welfare.
1c) Identify research priorities that have the most potential for further improving the practicality and effectiveness of on-farm welfare assessments.
2) Investigate how farm site compliance to welfare standards has changed over the years, and what insights this provides into the welfare practices of the Scottish salmon farming sector.
2a) Determine to what extent these welfare standards are able to provide evidence for what level of welfare has been achieved for farmed salmon.
2b) Investigate what limitations may exist for these standards, and why.
3) Validate the application of a novel welfare assessment tool that effectively addresses the key needs, highlighted from objectives 1-2, for further improving on-farm welfare assessment.

1.9.1 Project outline

Chapter 1 of this thesis consists of a comprehensive literature review covering key aspects of conceptualising animal welfare, the evidence for fish sentience and importance of farmed salmon welfare, factors and indicators relevant to farmed salmon welfare, and important considerations for on-farm welfare assessments.

Chapter 2 details a study which gathered opinions from the Scottish salmon farming industry in order to investigate the relative importance of different husbandry practices, production stages, overall welfare concerns, welfare indicators, and research priorities within the industry.

Chapter 3 investigates what role welfare standards have to play in providing assurances for farmed salmon welfare, as well as what role they could play in the future provided that improvements are made in the practicality of certain welfare indicators. In addition, this study also provides insights into how welfare practices within the industry have changed over the years through examining changes in farm site compliance to these standards.

Chapter 4 reports on the first study to demonstrate the ability of Qualitative Behavioural Assessment (QBA) to capture changes in the behavioural expression of Atlantic salmon following exposure to a stressful challenge. In this study, QBA was also correlated against other welfare measures (feed intake and darting behaviours) to further explore what role QBA could have as a welfare indicator.

Chapter 5 is a general discussion, highlighting how the findings from chapters 2 and 3 informed the development of the QBA experiment conducted in chapter 4. In addition, the overall outcomes of this PhD study are outlined. Finally, a direction for future research is proposed, regarding the potential for behavioural welfare assessment tools to utilise emerging technologies to further leverage the benefits of implementing non-intrusive, animal-based welfare indicators that can be carried out remotely.

1.10 References

Algers, B. et al. (2009) Scientific opinion - general approach to fish welfare and to the concept of sentience in fish Scientific Opinion of the Panel on Animal Health and Welfare PANEL MEMBERS. The EFSA Journal, 954(954), pp. 1–27.

Amoroso, G. et al. (2016) Multigenic delineation of lower jaw deformity in triploid Atlantic Salmon (*Salmo salar L.*). PLoS ONE, 11(12), pp. 1–21. doi: 10.1371/journal.pone.0168454.

Animal Welfare Act 2006 (2006) Animal Welfare Act 2006. The National Archives. Available at: https://www.legislation.gov.uk/ukpga/2006/45/contents (Accessed: 27 June 2022).

ASC (2019) ASC Salmon Standard Version 1.3. Available at: https://www.asc-aqua.org/what-we-do/our-standards/farm-standards/the-salmon-standard/ (Accessed: 29 January 2020).

Ashley, P. J. (2007) Fish welfare: Current issues in aquaculture. Applied Animal Behaviour Science, 104(3–4), pp. 199–235. doi: 10.1016/j.applanim.2006.09.001.

Barreto, M. O. et al. (2021) Emerging indicators of fish welfare in aquaculture. Reviews in Aquaculture, (December 2020), pp. 1–19. doi: 10.1111/raq.12601.

Bass, S. L. S. and Gerlai, R. (2008) Zebrafish (Danio rerio) responds differentially to stimulus fish : The effects of sympatric and allopatric predators and harmless fish, Behavioural Brain Research, 186, pp. 107–117. doi: 10.1016/j.bbr.2007.07.037.

Berg, A. G. T., Haagensen, E. and Horsberg, T. E. (2012) Anesthesia during vaccination of Atlantic salmon: Compliance with Recommended Procedures. Journal of the World Aquaculture Society, 43(1), pp. 120–127. doi: 10.1111/j.1749-7345.2011.00546.x.

Bergqvist, J. and Gunnarsson, S. (2013) Finfish Aquaculture: Animal Welfare, the Environment, and Ethical implications. Journal of Agricultural and Environmental Ethics, 26(1), pp. 75–99. doi: 10.1007/s10806-011-9346-y.

Botreau, R. et al. (2007) Definition of criteria for overall assessment of animal welfare. Animal Welfare, 16(2), pp. 225–228.

Braithwaite, V. A. and Huntingford, F. A. (2004) Fish and welfare: do fish have the capacity for pain perception and suffering?. Animal Welfare, 13, pp. 87–92.

Braithwaite, V. A., Huntingford, F. A. and Van den Bos, R. (2013) Variation in Emotion and Cognition Among Fishes. Journal of Agricultural and Environmental Ethics, 26(1), pp. 7–23. doi: 10.1007/s10806-011-9355-x.

Brambell, F. W. R. et al. (1965) Report of the Technical Committee to Enquire in to the Welfare of Animals kept under Intensive Livestock Husbandry Systems. London: HMSO.

Broom, D. M. (1991) Animal Welfare: concepts and measurement. Journal of Animal Science, 69(1), pp. 4167–4175.

Broom, D. M. (1999) Fish Welfare and the public perception of farmed fish. Proceedings of Aquavision. 98, pp. 89-91.

Broom, D. M. (2007) Cognitive ability and sentience: Which aquatic animals should be protected ?. Diseases of Aquatic Organisms, 75, pp. 99–108.

Brown, C. (2015) Fish intelligence, sentience and ethics. Animal Cognition, 18(1), pp. 1–17. doi: 10.1007/s10071-014-0761-0.

Brown, C., Davidson, T. and Laland, K. (2003) Environmental enrichment and prior experience of live prey improve foraging behaviour in hatchery-reared Atlantic salmon. Journal Of Fish Biology, 63(Supplement A), pp. 187–196. doi: 10.1017/9781316492390.

Brown, D. M. et al. (2008) Animal welfare aspects of husbandry systems for farmed Atlantic salmon - Scientific Opinion of the Panel on Animal Health and Welfare. The EFSA Journal, 6(7), pp. 736. doi: 10.2903/j.efsa.2008.796.

Browning, H. (2023) Improving welfare assessment in aquaculture. Frontiers in Veterinary Science, 10, pp. 1–10. doi: 10.3389/fvets.2023.1060720.

Cañon Jones, H. A. et al. (2010) Social network analysis of behavioural interactions influencing fin damage development in Atlantic salmon (*Salmo salar*) during feed-restriction. Applied Animal Behaviour Science, 127(3–4), pp. 139–151. doi: 10.1016/j.applanim.2010.09.004.

Chandroo, K. P., Duncan, I. J. H. and Moccia, R. D. (2004) Can fish suffer?: Perspectives on sentience, pain, fear and stress. Applied Animal Behaviour Science, 86(3–4), pp. 225–250. doi: 10.1016/j.applanim.2004.02.004.

Chervova, L. S. and Lapshin, D. N. (2011) Behavioral control of the efficiency of pharmacological anesthesia in Fish. Journal of Ichthyology, 51(11), pp. 1126–1132. doi: 10.1134/S0032945211110026.

Conte, F. S. (2004) Stress and the welfare of cultured fish. Applied Animal Behaviour Science, 86(3–4), pp. 205–223. doi: 10.1016/j.applanim.2004.02.003.

Cooke, M. (2017) An introduction to animal welfare issues in aquaculture. The Business of Farm Animal Welfare, pp. 272–285. Routledge. doi: 10.4324/9781351270045.

Corder, G., Ahanonu, B., Grewe, B. F. et al. (2019) An amygdalar neural ensemble that encodes the unpleasantness of pain. Science, 363(6424), pp. 276–281. doi: 10.1126/science.aap8586.

Council of the European Union (1998) Council Directive 98/58/EC concerning the protection of animals kept for farming purposes. Official Journal of the European Communities, 221(23), pp. 23–27. Available at: https://eur-lex.europa.eu/legal-content/EN/TXT/HTML/?uri=CELEX:31998L0058&from=EN.

Dawkins, M. S. (1990) From an animal’s point of view: Motivation, fitness, and animal welfare. Behavioral and Brain Sciences, 13, pp. 1–61.

Dawkins, M. S. (1998) Evolution and animal welfare. The Quarterly Review of Biology, 73(3), pp. 305–328.

Dawkins, M. S. (2001) Who needs consciousness?. Animal Welfare, 10(SUPPL.), pp. 19–29.

Dawkins, M. S. (2004) Using behaviour to assess animal welfare. Animal Welfare, 13(SUPPL.), pp. 3–7.

Dawkins, M. S. (2015) Animal welfare and the paradox of animal consciousness. Advances in the Study of Behavior. Elsevier Ltd, 47, pp. 5-38. doi: 10.1016/bs.asb.2014.11.001.

Ellis, T. et al. (2012) Mortality and fish welfare. Fish Physiology and Biochemistry, 38(1), pp. 189–199. doi: 10.1007/s10695-011-9547-3.

Ellis, T. et al. (2016) Trends during development of Scottish salmon farming: An example of sustainable intensification?. Aquaculture, Elsevier B.V., 458, pp. 82–99. doi: 10.1016/j.aquaculture.2016.02.012.

Ersdal, C., Midtlyng, P. J. and Jarp, J. (2001) An epidemiological study of cataracts in seawater farmed Atlantic salmon *Salmo salar*. Diseases of Aquatic Organisms, 45, pp. 229–236.

European Commission (2016) Attitudes of Europeans towards Animal Welfare - Special Eurobarometer. Available at: https://europa.eu/eurobarometer/surveys/detail/2096 (Accessed 10 January 2023).

FAO (2022) Global fisheries and aquaculture at a glance, The state of world fisheries and aquaculture 2022. Available at: https://www.fao.org/3/cc0461en/online/sofia/2022/aquaculture-production.html (Accessed: 11 October 2021).

Farrell, A. P., Johansen, J. A. and Suarez, R. K. (1991) Effects of exercise-training on cardiac performance and muscle enzymes in rainbow trout, *Oncorhynchus mykiss*. Fish Physiology and Biochemistry, 9(4), pp. 303–312. doi: 10.1007/BF02265151.

FAWC (2014) Opinion on the Welfare of Farmed Fish. *FAWC report,* pp. 1-40. Available at: https://assets.publishing.service.gov.uk/media/5a7eb32340f0b62305b829c4/Opinion_on_the_welfare_of_farmed_fish_at_the_time_of_killing.pdf (Accessed: 11 October 2021).

Fernö, A. and Holm, M. (1986) Aggression and growth of Atlantic Salmon parr. I. Different stocking densities and size groups. Fiskeridirektoratets Skrifter, Serie Havundersøkelser, 18(February), pp. 113–122.

Fife-Cook, I. and Franks, B. (2019) Positive welfare for fishes: rationale and areas for future study. Fishes, 4(2), p. 31. doi: 10.3390/fishes4020031.

Franks, B., Ewell, C. and Jacquet, J. (2021) Animal welfare risks of global aquaculture. Science Advances, 7, pp. 1–8.

Fraser, D. et al. (1997) A scientific conception of animal welfare that reflects ethical concerns. Animal Welfare, 6(3), pp. 1–19.

Fraser, D. (2003) Assessing animal welfare at the farm and group level: The interplay of science and values. Animal Welfare, 12(4), pp. 433–443.

Fraser, D. (2008) Understanding Animal Welfare. Acta Veterinaria Scandinavica, 50, pp. 1–7. doi: 10.1186/1751-0147-50-S1-S1.

Fraser, D. (2009) Assessing animal welfare: Different philosophies, different scientific approaches. Zoo Biology, 28(6), pp. 507–518. doi: 10.1002/zoo.20253.

FSBI (2002) Fish Welfare, Briefing Paper 2, Fisheries Society of the British Isles. Fish & Shellfish Immunology. doi: 10.1006/fsim.1999.0251.

Giménez-Candela, M., Saraiva, J. L. and Bauer, H. (2020) The legal protection of farmed fish in Europe – Analysing the range of EU legislation and the impact of international animal welfare standards for the fishes in European aquaculture. Derecho Animal, 11(1), pp. 65–119. doi: 10.5565/rev/da.460.

Global Aquaculture Alliance (2016) Salmon Farms B.A.P. Certification Standards. Available at: https://www.bapcertification.org/Standards (Accessed: 29 January 2020).

GlobalG.A.P. (2017) The GLOBALG.A.P. Integrated Farm Assurance (IFA) Standard. Available at: https://www.globalgap.org/uk_en/documents/#fq=gg.document.type:(%22checklist%22+OR+%22regulations%22+OR+%22cpacc%22)&fq=gg.standard.gg:(%22ifa5%22)&fq=gg.subscope:(%22aquacullture%22)&fq=con_locales:(%22en%22) (Accessed: 29 January 2020).

Green, T. C. and Mellor, D. J. (2011) Extending ideas about animal welfare assessment to include “quality of life” and related concepts. New Zealand Veterinary Journal, 59(6), pp. 263–271. doi: 10.1080/00480169.2011.610283.

Håstein, T., Scarfe, A. D. and Lund, V. L. (2005) Science-based assessment of welfare: Aquatic animals. OIE Revue Scientifique et Technique, 24(2), pp. 529–547. doi: 10.20506/rst.24.2.1590.

Hemsworth, P. H. et al. (2015) Scientific assessment of animal welfare. New Zealand Veterinary Journal, 63(1), pp. 24–30. doi: 10.1080/00480169.2014.966167.

Huntingford, F. A. et al. (2006) Current issues in fish welfare. Journal of Fish Biology, 68(January), pp. 332–372. doi: 10.1111/j.1095-8649.2005.01046.x.

Huntingford, F. A. and Kadri, S. (2009) Taking account of fish welfare: Lessons from aquaculture. Journal of Fish Biology, 75(10), pp. 2862–2867. doi: 10.1111/j.1095-8649.2009.02465.x.

Jarvis, S. et al. (2021) Qualitative behavioral assessment in juvenile farmed Atlantic salmon (Salmo salar): Potential for On-Farm Welfare Assessment. Frontiers in Veterinary Science, 8(September), pp. 1–11. doi: 10.3389/fvets.2021.702783.

Jones, R. C. (2013) Science, sentience, and animal welfare. Biology and Philosophy, 28(1), pp. 1–30. doi: 10.1007/s10539-012-9351-1.

Juell, J. E. et al. (2003) Submerged light increases swimming depth and reduces fish density of Atlantic salmon *Salmo salar L.* in production cages. Aquaculture Research, 34(6), pp. 469–477. doi: 10.1046/j.1365-2109.2003.00833.x.

Korte, S. M., Olivier, B. and Koolhaas, J. M. (2007) A new animal welfare concept based on allostasis. Physiology and Behavior, 92(3), pp. 422–428. doi: 10.1016/j.physbeh.2006.10.018.

Marschall, E. A. et al. (1998) A framework for understanding Atlantic salmon (Salmo salar) life history. Canadian Journal of Fisheries and Aquatic Sciences, 55(SUPPL.1), pp. 48–58. doi: 10.1139/cjfas-55-s1-48.

McGregor, P. K., Peake, T. M. and Lampe, H. M. (2001) Fighting fish Betta splendens extract relative information from apparent interactions: What happens when what you see is not what you get. Animal Behaviour, 62(6), pp. 1059–1065. doi: 10.1006/anbe.2001.1850.

McNeill, B. and Perry, S. F. (2006) The interactive effects of hypoxia and nitric oxide on catecholamine secretion in rainbow trout (*Oncorhynchus mykiss*). Journal of Experimental Biology, 209(21), pp. 4214–4223. doi: 10.1242/jeb.02519.

Mellor, D. J. (2012) Animal emotions, behaviour and the promotion of positive welfare states. New Zealand Veterinary Journal, 60(1), pp. 1–8. doi: 10.1080/00480169.2011.619047.

Mellor, D. J. (2016) Updating animal welfare thinking: moving beyond the “five freedoms” towards “A life worth living”. Animals, 6(3), pp. 1–20. doi: 10.3390/ani6030021.

Mettam, J. J. et al. (2011) The efficacy of three types of analgesic drugs in reducing pain in the rainbow trout, *Oncorhynchus mykiss*. Applied Animal Behaviour Science. Elsevier B.V., 133(3–4), pp. 265–274. doi: 10.1016/j.applanim.2011.06.009.

Morton, D. B. and Griffiths, P. H. (1985) Guidelines on the recognition of pain, distress and discomfort in experimental animals and an hypothesis for assessment. The Veterinary record, 116(16), pp. 431–436. doi: 10.1136/vr.116.16.431.

Munro, L. A. (2022) Scottish Fish Farm Production Survey 2021. Available at: https://www.gov.scot/publications/scottish-fish-farm-production-survey-2021/pages/5/. Accessed: 30th October, 2019.

Näslund, J. et al. (2013) Hatchery tank enrichment affects cortisol levels and shelter-seeking in Atlantic salmon (*Salmo salar*). Canadian Journal of Fisheries and Aquatic Sciences, 70(4), pp. 585–590. doi: 10.1139/cjfas-2012-0302.

Noble, C. et al. (2012) Injuries and deformities in fish: their potential impacts upon aquacultural production and welfare. Fish Physiology and Biochemistry, 38, pp. 61–83. doi: 10.1007/s10695-011-9557-1.

Noble, C., Gismervik, K., Iversen, M. H. et al. (eds.) (2018) Welfare Indicators for farmed Atlantic Salmon: tools for assessing fish welfare http://hdl.handle.net/11250/2575780.

O'Connor, K. I., Metcalfe, N. B. and Taylor, A. C. (2000) Familiarity influences body darkening in territorial disputes between juvenile salmon. Animal Behaviour, 59(6), pp. 1095–1101. doi: 10.1006/anbe.2000.1401.

Ohl, F. and Van der Staay, F. J. (2012) Animal welfare: At the interface between science and society. Veterinary Journal, 192(1), pp. 13–19. doi: 10.1016/j.tvjl.2011.05.019.

Oliva-Teles, A. (2012) Nutrition and health of aquaculture fish. Journal of Fish Diseases, 35(2), pp. 83–108. doi: 10.1111/j.1365-2761.2011.01333.x.

Oliveira, R. F., McGregor, P. K. and Latruffe, C. (1998) Know thine enemy: Fighting fish gather information from observing conspecific interactions. Proceedings of the Royal Society B: Biological Sciences, 265(1401), pp. 1045–1049. doi: 10.1098/rspb.1998.0397.

Oppedal, F., Dempster, T. and Stien, L. H. (2011) Environmental drivers of Atlantic salmon behaviour in sea-cages: A review. Aquaculture, 311(1–4), pp. 1–18. doi: 10.1016/j.aquaculture.2010.11.020.

Oppedal, F., Juell, J. E. and Johansson, D. (2007) Thermo- and photoregulatory swimming behaviour of caged Atlantic salmon: Implications for photoperiod management and fish welfare. Aquaculture, 265(1–4), pp. 70–81. doi: 10.1016/j.aquaculture.2007.01.050.

Panksepp, J. (2005) Affective consciousness: Core emotional feelings in animals and humans. Consciousness and Cognition, 14, pp. 30–80. doi: 10.1016/j.concog.2004.10.004.

Poppe, T. T., Barnes, A. C. and Midtlyng, P. J. (2002) Welfare and ethics in fish farming, Bulletin of the European Association of Fish Pathologists. Bulletin-European Association of fish pathologists, 22(2), pp. 148–151.

Powell, M. D., Reynolds, P. and Kristensen, T. (2015) Freshwater treatment of amoebic gill disease and sea-lice in seawater salmon production: Considerations of water chemistry and fish welfare in Norway. Aquaculture, 448, pp. 18–28. doi: 10.1016/j.aquaculture.2015.05.027.

Remen, M. (2012) The oxygen requirement of Atlantic salmon (*Salmo salar L.*) in the on-growing phase in sea cages. Doctoral thesis. Available at: http://bora.uib.no/bitstream/handle/1956/9535/dr.thesis_Mette_Remen_30102012.pdf?sequence=3&isAllowed=y Accessed October 30th, 2019.

Rey Planellas, S., Little, D. and Ellis, M. (2019) Farmed fish welfare practices: salmon farming as a case study, GAA publications.

Robertsen, B. (2011) Can we get the upper hand on viral diseases in aquaculture of Atlantic salmon?. Aquaculture Research, 42(SUPPL. 1), pp. 125–131. doi: 10.1111/j.1365-2109.2010.02671.x.

Rose, J. D. (2002) The neurobehavioral nature of fishes and the question of awareness and pain. Reviews in Fisheries Science, 10(1), pp. 1–38. doi: 10.1080/20026491051668.

Rose, J. D. et al. (2014) Can fish really feel pain?. Fish and Fisheries, 15(1), pp. 97–133. doi: 10.1111/faf.12010.

RSPCA (2018) RSPCA welfare standards for farmed Atlantic salmon. Available at: https://science.rspca.org.uk/sciencegroup/farmanimals/standards/salmon (Accessed: 28 October 2019).

RSPCA (2021) RSPCA welfare standards for Farmed Atlantic Salmon, RSPCA welfare standards for Farmed Atlantic Salmon, (February), pp. 1–80. Available at: http://industry.freedomfood.co.uk/media/96636/RSPCA-welfare-standards-for-farmed-Atlantic-salmon-web-version.pdf.

Salmon Scotland (2020) What is RSPCA Assured?. Fish Health & Welfare. Available at: https://www.salmonscotland.co.uk/facts/fish-health-welfare/what-is-rspca-assured (Accessed: 11 January 2023).

Santurtun, E., Broom, D. M. and Phillips, C. J. C. (2018) A review of factors affecting the welfare of Atlantic salmon (*Salmo salar*). Animal Welfare, 27(3), pp. 193–204. doi: 10.7120/09627286.27.3.193.

Scottish Parliament (2006) Animal Health and Welfare (Scotland) Act 2006. Acts of the Scottish Parliament. Available at: https://www.legislation.gov.uk/asp/2006/11/contents (Accessed: 11 June 2020).

Scottish Salmon Producers Organisation (2015) Code of Good Practice for Scottish Finfish Aquaculture. Available at: http://thecodeofgoodpractice.co.uk/chapters/ (Accessed: 15 November 2019).

Silverstone, A. M. and Hammell, L. (2002) Spinal deformities in farmed Atlantic salmon. Canadian Veterinary Journal, 43(10), pp. 782–784.

Sloman, K. A. et al. (2019) Ethical considerations in fish research. Journal of Fish Biology, 94(4), pp. 556–577. doi: 10.1111/jfb.13946.

Sneddon, L. U. (2003) The evidence for pain in fish: The use of morphine as an analgesic. Applied Animal Behaviour Science, 83(2), pp. 153–162. doi: 10.1016/S0168-1591(03)00113-8.

Sneddon, L. U. (2009) Pain perception in fish: Indicators and endpoints. ILAR Journal, 50(4), pp. 338–342. doi: 10.1093/ilar.50.4.338.

Sneddon, L. U. (2015) Pain in aquatic animals. Journal of Experimental Biology, 218(7), pp. 967–976. doi: 10.1242/jeb.088823.

Sneddon, L. U., Braithwaite, V. A. and Gentle, M. J. (2003) Do fishes have nociceptors? Evidence for the evolution of a vertebrate sensory system. Proceedings of the Royal Society B: Biological Sciences, 270(1520), pp. 1115–1121. doi: 10.1098/rspb.2003.2349.

Solstorm, F. et al. (2015) Fast water currents reduce production performance of post-smolt Atlantic salmon *Salmo salar*. Aquaculture Environment Interactions, 7(2), pp. 125–134. doi: 10.3354/aei00143.

Solstorm, F. et al. (2016) Not too slow, not too fast: Water currents affect group structure, aggression and welfare in post-smolt Atlantic salmon *Salmo salar*. Aquaculture Environment Interactions, 8, pp. 339–347. doi: 10.3354/aei00178.

Southgate, P. and Wall, T. (2001) Welfare of farmed fish at slaughter. In Practice, 23, pp. 277–284.

Stien, L. H. et al. (2013) Salmon Welfare Index Model (SWIM 1.0): A semantic model for overall welfare assessment of caged Atlantic salmon: Review of the selected welfare indicators and model presentation. Reviews in Aquaculture, 5(1), pp. 33–57. doi: 10.1111/j.1753-5131.2012.01083.x.

Størkersen, K. V. et al. (2021) Fish protection during fish production. Organizational conditions for fish welfare. Marine Policy, 129(October 2020), pp. 1–11. doi: 10.1016/j.marpol.2021.104530.

Taylor, J. G. (2001) What do neuronal network models of the mind indicate about animal consciousness?. Animal Welfare, 10(SUPPL.), pp. 63–75.

Thorstad, E. B. et al. (2011) Aquatic Nomads: The life and migrations of the Atlantic salmon. Atlantic salmon ecology, 1(6), pp. 1-32. doi: 10.1002/9781444327755.ch1.

Tidwell, J. H. and Allan, G. L. (2001) Fish as food: Aquaculture’s contribution. Ecological and economic impacts and contributions of fish farming and capture fisheries. EMBO Reports, 2(11), pp. 958–963. doi: 10.1093/embo-reports/kve236.

Turnbull, J. F. et al. (2005) Stocking density and welfare of cage farmed Atlantic salmon: Application of a multivariate analysis. Aquaculture, 243(1–4), pp. 121–132. doi: 10.1016/j.aquaculture.2004.09.022.

Turnbull, J. F. and Kadri, S. (2007) Safeguarding the many guises of farmed fish welfare. Diseases of Aquatic Organisms, 75(2), pp. 173–182. doi: 10.3354/dao075173.

Turnbull, J. F., Richards, R. H. and Robertson, D. A. (1996) Gross, histological and scanning electron microscopic appearance of dorsal fin rot in farmed Atlantic salmon, *Salmo salar L.*, parr. Journal of Fish Diseases, 19(6), pp. 415–427. doi: 10.1111/j.1365-2761.1996.tb00381.x.

UK Parliament (1974) Slaughterhouses Act 1974. Available at: https://www.legislation.gov.uk/ukpga/1974/3/enacted#:~:text=(1)Subject%20to%20subsection%20(,in%20force%20under%20this%20section. (Accessed: 30th October 2019).

UK Parliament (1995) The Welfare of Animals (Slaughter or Killing) Regulations 1995, UK Statutory Instruments. Available at: https://www.legislation.gov.uk/uksi/1995/731/contents/made (Accessed: 11 June 2020).

UK Parliament (2007) The Welfare of Farmed Animals (England) Regulations 2007, UK Statutory Instruments. Available at: https://www.legislation.gov.uk/uksi/2007/2078/contents/made (Accessed: 11 June 2020).

Voas, A. (2008) Welfare Legislation Applying to Farmed Fish in the UK, in Branson, E. J. (ed.) Fish Welfare. Oxford, UK: Blackwell Publishing Ltd, pp. 93–100.

Warburton, K. (2003) Learning of foraging skills by fish. Fish and Fisheries, 4(3), pp. 203–215. doi: 10.1046/j.1467-2979.2003.00125.x.

Webster, J. (2016) Animal welfare: Freedoms, dominions and “A life worth living”, Animals, 6(6), pp. 2–7. doi: 10.3390/ani6060035.

Yoshida, M. and Hirano, R. (2010) Effects of local anesthesia of the cerebellum on classical fear conditioning in goldfish. Behavioural and Brain Functions, 6(1), pp. 1–9.

CHAPTER 2. Concerns and research priorities for Scottish farmed salmon welfare – an industry perspective

Timothy Robert Wiese[1], Marie Haskell[2], Susan Jarvis[3], Sonia Rey Planellas[1], Jimmy Turnbull[1]

[1]: Institute of Aquaculture, University of Stirling, Stirling, FK9 4LA, UK.

[2]: SRUC (Scotland's Rural College),West Mains Road, Edinburgh, EH9 3JG, UK

[3]: Global Academy of Agriculture and Food Systems, University of Edinburgh, Midlothian, EH25 9RG, UK.

2.1 Abstract

The intensification of Scottish salmon farming has been associated with increasing demands for the monitoring and safeguarding of farmed salmon welfare. Continued growth of farm productivity, while avoiding adverse effects on salmon welfare, will require the development of effective welfare assessment tools. This paper reports on a survey of the Scottish salmon farming industry, which was conducted to understand current salmon welfare concerns and priorities for research. As part of a broader aim for further developing tools for on-farm salmon welfare assessment, a total of 61 individuals working in the Scottish salmon farming industry took part. This survey intentionally focused on industry stakeholders to provide insights into current practices and challenges associated with monitoring and assessing salmon welfare. Participants were recruited through authors' industry contacts, online advertisements, and searches of company websites. In terms of production stages, survey participants believed that the seawater rearing stage is a major area of concern, largely due to the challenges presented by sea lice. Gill health and environmental challenges, mainly relating to water quality, were two other highly ranked welfare concerns. Methods to monitor salmon welfare during husbandry practices, where disturbances and contact with the salmon is unavoidable (particularly during crowding, grading, and interventions), were emphasised as a priority. Although these were identified as the major concerns, the survey indicated that there are other significant welfare concerns specific to each production stage that also require consideration. Participants highlighted non-invasive, remote, and animal-based welfare measures as important areas for further development for on-farm welfare assessments. Behavioural measures were identified as having the potential to make a major contribution in this context. This survey presents the first collection of opinions from professionals employed across the Scottish salmon farming industry regarding the current overall state of farmed salmon welfare. This study upholds the importance of using an integrated approach to welfare assessments, and that behavioural measures could play an important role in ensuring these assessments benefit both salmon welfare and farm productivity.

Keywords:

Aquaculture; fish health; non-invasive monitoring; animal behaviour; survey.

2.2 Introduction

Farmed salmon welfare is inextricably linked to the farming practices and conditions within the salmon farming industry (FAWC, 2014; Noble *et al.*, 2018). Animal welfare encompasses the physical and emotional state of an animal, its ability to cope with external events, and its overall quality of life (as a cumulative result of those events) (Webster, 2016). Animal welfare is often now an important factor for the public when deciding whether a husbandry system's continued use is acceptable on ethical grounds (Broom, 2011). In the UK, farmed fish are also protected with a duty of care requirement under the Animal Welfare Act (2006), with the majority of salmon farms (~70%) also being certified by the RSPCA Assured standards (Salmon Scotland, 2020b). Additionally, stress and poor welfare are known to increase susceptibility to disease, increase mortality rates, and ultimately lead to poor production (Schreck and Tort, 2016). For reference, Scottish salmon farming generated a direct economic contribution of £468 million in gross value added in 2018 (Economics, 2020), placing Scotland as the third largest producer of Atlantic salmon in the world (Kenyon and Davies, 2018). Safeguarding salmon welfare should therefore be seen as a priority from a moral, economic, and legal perspective (Animal Welfare Act 2006; Lafferty *et al.*, 2015).

A detailed understanding of the current state of the industry, with regards to welfare, is then also required to make valid, industry-relevant contributions to farmed salmon welfare. This includes identifying current concerns facing farmed salmon welfare, along with having knowledge of relevant production stages, husbandry practices, and the practicalities of on-farm welfare monitoring and assessment. Such information plays a vital role in developing a framework for realistic improvements of welfare assessments that, when used, do not come at the cost of farm productivity.

Various frameworks have been designed to help form the basis of animal welfare management. The Five Domains model, developing upon the Five Freedoms (Mellor, 2016), was created to provide a more systematic method for identifying potential welfare impacts associated with events/situations (Mellor and Reid, 1994). These impacts were divided into four physical domains (nutrition, environment, health/functional status, behaviour) and one mental domain (overall mental state). Originally designed to assess "compromise" in the welfare state of an animal, recent extensions to the Five Domains now facilitate considerations of positive experiences that may enhance welfare (Mellor and Beausoleil, 2015). It is now widely accepted that emotional affected states are an essential consideration when promoting positive welfare (Dawkins, 2004, 2006; Fisher, 2009; Paul *et al.*, 2020). The development of welfare assessment tools should then not only focus on physical well-being and avoiding ‘negative’ welfare, but also promoting emotional well-being and ‘positive’ welfare (Fife-Cook and Franks, 2019).

In order to capture these different aspects of animal welfare, current welfare assessments typically include a set of ‘Operational Welfare Indicators’ (OWIs) that are believed to be practical and appropriate for detecting changes to the animal’s welfare status (Noble *et al.*, 2018). Examples of such welfare assessments include the monitoring program for physical damage or deformities suggested in the RSPCA Assured welfare standards for farmed Atlantic salmon (RSPCA, 2018b), and the Salmon Welfare Index Models (SWIM 1.0 and SWIM 2.0) (Stien *et al.*, 2013; Pettersen *et al.*, 2014). Selected OWIs range from environmental (e.g., water temperature, oxygen saturation, salinity) to animal-based indicators (e.g., fin/eye/snout damage, deformities, changes in behaviour, sea lice infestation) (Stien *et al.*, 2013; Noble *et al.*, 2018). As on-farm assessments are limited to including Welfare Indicators (WIs) that are currently practical and affordable to use (Noble *et al.*, 2018), it is likely that the full potential for how we can monitor and safeguard farmed salmon welfare is not yet realised. Stien et al. (2013) anticipate that these assessments, including their SWIM 1.0 model, will need further “upgrading” either through the development of current WIs or inclusion of entirely new WIs.

Identifying where improvements should be made, either within on-farm assessments or the general management of salmon welfare, is no easy feat. Monitoring and safeguarding farmed salmon welfare presents various challenges due to their complex, anadromous life cycle (Marschall *et al.*, 1998). There are rearing conditions, husbandry practices, responsibilities and welfare considerations that are specific to each production stage (Bergqvist and Gunnarsson, 2013; Noble *et al.*, 2018). The total tonnage of seawater fish produced per employee has also increased over 10-fold since 1985 (Ellis *et al.*, 2016). This increasing intensity of production means that work practices, including welfare assessment tools, have to be time-efficient in order to be practical in the commercial production environment. When forming opinions on welfare concerns within this context, on-site experience of the various production stages can provide important perspectives on both the current practices and the relevant challenges that are faced. Professionals employed in salmon farming may potentially have a better understanding of how the processes involved are linked with salmon welfare, particularly with what practical limitations there are when implementing welfare assessments into their farming routines. Including these considerations during any developments of welfare assessment tools or management therefore increases the likelihood of their adoption on-site. Production staff ultimately play an essential role in safeguarding salmon welfare, where they share knowledge and develop and execute routines to protect farmed fish (Størkersen *et al.*, 2021). In this survey, we attempted to access the collective knowledge of these production staff with the assumption that it would provide valuable insights into where farmed salmon welfare can be further advanced.

To date, no study has been conducted which focuses solely on professionals directly employed in Scottish salmon farming to assess their opinions on the state of this industry with regards to salmon welfare. A broader gap-analysis study was carried out on stakeholders from the European aquaculture sector and research community in 2018, with an aim of investigating research priorities for overall farmed fish welfare (Manfrin, Messori and Arcangel, 2018). However, in this 2018 study, Atlantic salmon were just one of nine species investigated over several countries.

It has been suggested that taking into account different perspectives within a particular industry, and working towards building a clear consensus on future research priorities, provides the best foundation for progressing fish welfare (Manfrin, Messori and Arcangel, 2018). This approach should be no different when making progress in the monitoring and management of farmed salmon welfare. This could take the form of farm staff either helping to identify key areas of concern, or highlighting any considerations that need to be made when improving on-farm assessments or the overall management of salmon welfare. Hence, this survey aimed to answer the following research questions:

1) Investigate the relative importance that different production stages, husbandry practices, and specific welfare concerns have towards farmed salmon welfare, as perceived by farm staff.
1a) In addition, assess any potential differences in these opinions and perceptions between farm staff with different professional backgrounds.
2) Identify which research priorities have the most potential for further improving the practicality and efficacy of on-farm salmon welfare measures.

Through addressing these research questions, this study will have provided a substantial contribution towards developing the practicality and efficacy of on-farm welfare assessment and management.

2.3 Materials and methods

2.3.1 Recruitment and survey development

Ethical approval for the survey development, recruitment methods and final version of the survey was obtained from the General University Ethics Panel (GUEP) at the University of Stirling (Project identification code GUEP (19 20) 858).

Survey development began with a key informant interview with two staff from a local hatchery. The discussion was based on open-ended questions regarding salmon welfare, prepared in advance of the interviews. These questions acted as a starting point for discussing general welfare concerns, which allowed the first version of the survey questions to be drafted. The first survey draft was piloted on 10 volunteers across different farming companies during a fish welfare course delivered at the Institute of Aquaculture, Stirling (February 2020). With each iteration of the survey, the following feedback (based on either responses to the survey questions or post-interview discussions) was obtained from participants to help further refine the questions and survey structure:

- For any of the question sections, was there any relevant information, topics, or opinions that participants felt were important to still share but they did not get the chance to?
- Were there any important questions or topics left out of the survey that participants felt were missing?
- Did the wording or structuring of any questions confuse participants in any way? I.e., was every question concise and easy to understand, and if not, why?
- Did the responses gathered for each question section provide valuable, relevant insights that help with your research objectives?
- Did the structuring / style of question allow for responses to be compared and assessed in a quantifiable manner (i.e., did they allow for inferential statistics to be carried out on them if that was the original goal)?

Responses and feedback from this first draft were gathered alongside a concurrent literature review, which focused partly on welfare assessment and factors influencing farmed salmon welfare to refine the focus of the final survey. Following this first draft, these initial research objectives were formulated for the survey:

- Determine the perceived importance of monitoring salmon welfare in the various production stages.
- Identify major areas of welfare concerns affecting farmed salmon.
- Identify which husbandry practices require the most attention to monitor and safeguard salmon welfare.
- Determine the practicality and efficacy for on-farm use of welfare measures.
- Determine salmon welfare research priorities.
- Identify which farming practices provide suitable opportunities for monitoring salmon welfare.

A second draft, modified on the basis of these research questions, was developed and piloted with volunteers at the Institute of Aquaculture, along with several key informants in the industry (n=7). This 2nd draft was piloted through in-person interviews and online formats (Microsoft Forms) to assess the effectiveness of the different styles and estimate the time for completion. Statistical analysis was not appropriate due to the small sample size, but essential feedback was gathered resulting in further refinement of the survey design.

2.3.2 Final questionnaire design

The final questionnaire, consisting of 53 questions, was divided into a section on participant's background followed by six question sections (see A1 in appendix). Background variables of participants (experience of specific production stage in salmon farming, current job title, and total years of experience in salmon farming) were recorded. Participants were informed about data security, and that any information they provided would remain anonymous. Due to the length of the questionnaire and inclusion of open-ended responses, constant and explicit signposts were used to emphasize the aim of each question section and prevent participants from drifting in their focus.

Section 1 asked participants to compare the relative importance of monitoring salmon welfare across the various production stages. Participants were provided with a list of the different production stages and asked to score each stage on a scale of 1 to 5 in terms of importance (1 = most important, 5 = least important).

Section 2 investigated the major areas of concern facing overall farmed salmon welfare. Section 3 examined which husbandry practices, due to their potential impacts, required the most attention towards monitoring salmon welfare. For these two sections, participants were asked to provide a minimum of three of their own examples in order of importance.

Section 4 examined what welfare measures were deemed most appropriate for on-farm use. Participants rated a list of welfare measures, on a scale of 1 to 10, by their practicality and effectiveness (1 = completely impractical / ineffective, 5-6 = somewhat practical / effective, 10 = very practical/effective). 'Practicality' was defined as 'how easy the measure is to use on-site', and 'effectiveness' was defined as 'how much valuable information the measure provides regarding the welfare status of the salmon'. Alongside each of these measures, participants were able to provide open-ended comments regarding any practical considerations that should be involved with the on-farm use of these measures. For the purpose of this paper, the term 'welfare measure' merely denotes a certain approach to assessing welfare, and is synonymous to 'welfare indicator'.

Section 5 asked participants to rate a list of research priorities, on a scale of 1 to 10, by the relevance and urgency of their development for on-farm welfare monitoring and assessment (1 = completely irrelevant / Not urgent at all, 5-6 = somewhat relevant / urgent, 10 = extremely relevant / urgent). 'Relevance' was defined as 'How relevant the need is for developing this group of welfare measures to allow for better monitoring and safeguarding of salmon welfare', whereas 'urgency' was defined as 'To what degree does this group of welfare measures need to be developed as soon as possible?'.

Section 6 explored which parts of a salmon farmer's daily routine provide the best opportunity for monitoring salmon welfare. Participants were able to select a maximum of three husbandry routines from a list of 5 (feeding times, health checks, routine inspections, grading and/or transfer, during video monitoring) as well as add their own response in free text.

Participation was voluntary through an online version of the survey through Microsoft Forms. As of 2020, 1,651 staff have been employed in Scottish salmon production (Munro, 2020). Efforts were made to ensure that as many of these staff as possible were at least informed of the opportunity to participate. This process began with colleagues forwarding the survey to potential participants, along with an introductory letter explaining the purpose of the survey. Advertisements and articles for the survey were shared across multiple media outlets, including fish farming news websites, Twitter and Facebook pages, community forums, accreditation sites, and company newsletters. A number of major Scottish salmon farming companies also agreed to support recruitment by forwarding the survey through their mailing lists. Individuals were also recruited directly via LinkedIn. The final survey was conducted from March-December 2020, where a total of 61 individuals directly employed within Scottish salmon production were consulted. Individuals who participated in the pilot studies were not included in the main survey.

2.3.3 Data processing and analysis

Data from the online survey were consolidated into Microsoft Excel (2019), where figures were also produced. Statistical analysis was then carried out using IBM SPSS Statistics 28 for Windows 10.

2.3.3.1 Quantitative responses

For section 1, weighted scores were created to reflect participants' rankings, which gave more weight to participants' scores indicating a higher priority (e.g., each score of "1" = 5 points, score of "2" = 4 points, "3" = 3 points, and so on). Total weighted scores were then calculated for each production stage. For sections 2 and 3, responses encompassing the same topic of welfare concern or husbandry practice were compiled into categories to allow comparisons to be made between these categories (see Table A1 and A2 in the appendix for a breakdown of these categories). For example, welfare concerns that included “*AGD*”, “*Gill Disease*”, and “*Gill Problems*” were placed into the category “*Gill Health*”. Husbandry practices that included “*Treatments*”, “*Mechanical / Chemical / Medicinal Treatments*”, and “*Vaccinations*” were placed into the category “*Interventions*”. The category *‘Handling’* included husbandry practices such as ‘*Crowding’, ‘Grading’, and ‘(Physical) Handling’*. Welfare concerns that included “Water quality” or “Environmental challenges” formed most of the category ‘Environmental challenges’. However, a minority of more specific concerns such as “Tidal throughput”, “Water temperature”, and “Climate change effects on SW” were also included in this category. Weighted scores were then calculated for each category of responses in the same manner as section 1. For the open-ended responses in sections 2 and 3, weighted scores helped ensure that the order/priority of participants’ responses would further reflect their significance, rather than assessing solely by the frequency of mentions. This would help distinguish categories that would have been referred to the same number of times, but at different “rankings” (first vs. last).

For the quantitative responses in Sections 1, 4 and 5, normality and homogeneity of variance were assessed before any parametric statistical analyses could be carried out. Log transformations were carried out on data sets to meet statistical assumptions when appropriate, but the degree of skewness for each data set (question sections 1, 4, and 5) did not allow for parametric tests. Therefore, Friedman’s tests and Kruskal Wallis tests were used on ordinal and interval data sets respectively to test for significant differences between the categories of responses. Where appropriate (where $p<0.05$), their corresponding post-hoc tests (Wilcoxon signed ranks test and Pairwise comparisons respectively) were then carried out with a Bonferroni correction. This allowed an assessment to identify where any statistically significant differences lay between categories of responses.

2.3.3.2 Qualitative responses

Open-text comments, regarding what practical considerations there are for implementing the specified measures on-site, were first input into excel and categorised by the welfare measure the comments were referring to. Within the collective raw text of comments associated with each welfare measure, recurring words, phrases, and topics were first identified with the use of word clouds. This helped to categorise certain words or phrases into ‘sub-themes’, which were essentially specific, recurring costs or benefits associated with using each welfare measure as mentioned by participants. For example, ‘early warning sign’ was identified as a recurring phrase mentioned across multiple welfare measures, and was therefore selected as one of the 25 sub-themes for this thematic analysis. The raw text was then input into Nvivo qualitative data analysis software (QSR International Pty Ltd., 2020). Each of these sub-themes (i.e., a specific type of cost or benefit associated with using the welfare measure) could then be coded in Nvivo as a “node”. The use of nodes allowed Nvivo to link each raw text comment to a certain sub-theme anytime the related key word or phrase corresponding to the sub-theme was mentioned. This consequently provided a frequency for the number of times each sub-theme was mentioned for each welfare measure. Based on the similarity of costs or benefits mentioned (i.e., whether they impacted or benefitted farm practices / salmon welfare), all 25 sub-themes were then grouped into five general themes. These were ‘Advantages to using welfare measure’, ‘Practicalities regarding use of equipment & facilities’, ‘Limitations to using welfare measure effectively’, ‘Practical limitations to using welfare measure on-site’, and ‘Negative impacts of using welfare measure’. For example, the theme ‘Advantages to using welfare measure’ included the sub-themes ‘early warning sign’ and ‘already taken as part of farm routine’. The theme ‘Limitations to using welfare measure effectively’ included sub-themes such as ‘ensuring representative sample size’ and ‘inherently subjective to score or notice’.

Within Nvivo, separate matrix queries were then carried out against the raw text of participants’ comments for each group of welfare measures; this quantified the frequency that each sub-theme / theme was mentioned for each group of welfare measures (e.g., across all participants’ comments, there were x amount of comments mentioning practical limitations to using this measure on-site). The frequency of themes mentioned for each group of welfare measures then helped with comparing the general sentiment of practicality involved between using the different welfare measures on-site.

2.3.3.3 Relationship between participants’ professional backgrounds and their responses

Where there was no clear consensus in responses across all participants, we assessed whether any difference in responses were significantly correlated with participant’s professional backgrounds.

For question sections 2, 3, and 6, participants were allowed to list and rank their own open-ended responses. Due to the lack of uniformity in the type of responses between participants, it was not possible to analyse the relationships between responses and backgrounds. Instead, these responses were examined separately for the different cohorts.

For question sections 1, 4, and 5, the homogeneity of responses/ratings between participants allowed for General Linear Models (GLM) to be used to examine potential relationships between the participant’s background and the responses they provided. Separate GLMs were carried out for each background variable (specific production experience, current job title, or years of salmon farming experience) and the responses within each question section. Ratings and background variables were included as fixed factors. To avoid pseudo replication in the GLM tests, participant ID numbers were included as a random effect.

2.4 Results

2.4.1 Key characteristics of participants

There was considerable diversity between the 61 participants' professional backgrounds (see Figure 2-1). Participants ranged from farmer trainees to production directors, with almost 50% of participants consisting of farm managers. Total on-farm experience ranged from <1 to 39 years, with an average of 14.5 years and more than half of the participants having more than a decade of experience in salmon farming. The majority of participants (82%) had some form of experience in the seawater rearing stage, whereas only 57% of participants had some form of freshwater experience. Where GLMs could be carried out, no relationship was found between these background variables (current position, years of experience, and production stage-specific experience) and the participants' responses ($p > 0.05$). Because of this, most question sections are described below with the responses from different cohorts combined. In certain question sections, not all participants provided answers (or at least provided responses that were relevant to topic in question) and as a result were removed from consideration. This is reflected in the number provided within each relevant figure (e.g., ‘n = x’, where x is the number of participants that provided relevant responses).

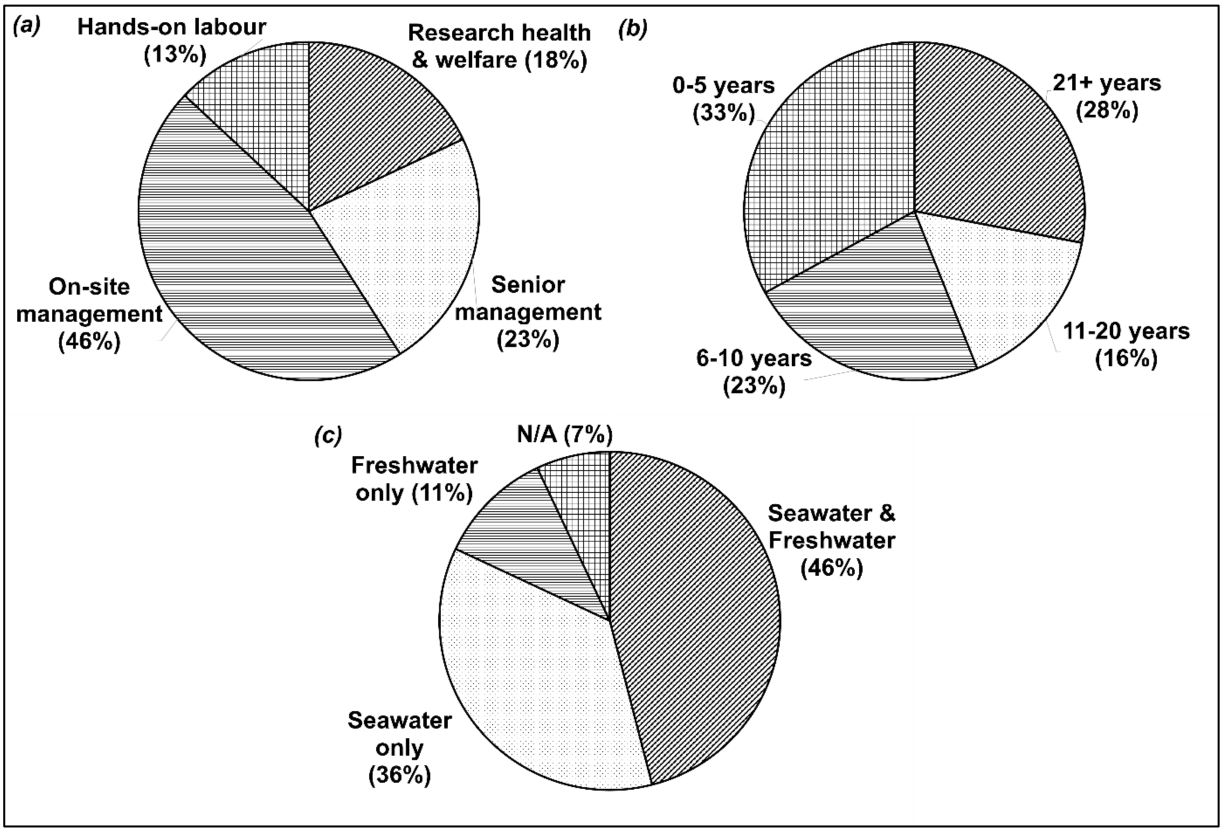

Figure 2-1. Breakdown of participants’ (n=61) professional backgrounds, including (a) their current job title, (b) total years of experience in salmon farming, and (c) what specific experience they have had across the different production stages. Participants were categorised into one of the four different groups for each of the three different background factors recorded.

2.4.2 Section 1 – Production stages; relative importance for monitoring salmon welfare

The seawater rearing stage received the highest numerical weighted score of relative importance. Significant differences in the Friedman test were also found between some of these weighted scores ($\chi^2 = 10.25$, df = 3, $P < 0.05$, see Figure 2-2). Seawater rearing and smoltification received comparable weighted scores. Although there were significant differences found between certain production stages, no single stage scored significantly different from all 3 other stages.

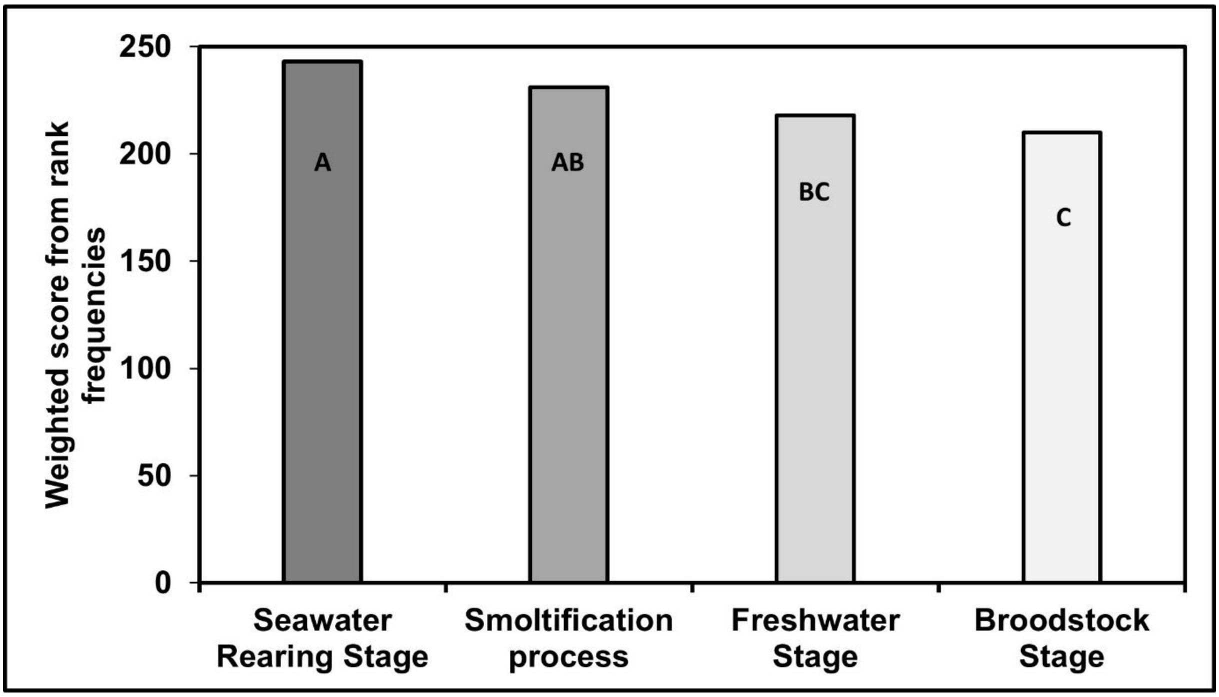

Figure 2-2. Relative importance of monitoring and assessing salmon welfare during each production stage, based on weighted scores provided by participants (n=61). Production stages without matching letters indicate a statistical difference ($P < 0.05$).

2.4.3 Section 2 – Overall farmed salmon welfare concerns

Out of the 10 highest scoring categories of welfare concerns listed (see Figure 2-3), 55% of the total weighted score was accounted for by the top 3 scoring categories ('Sea lice', 'Gill health', and 'Environmental challenges'). When listing 'sea lice' as a concern, 9 participants specifically referred to treatments for sea lice as one of their largest overall welfare concerns. A significant drop in the weighted scores followed, with 'Interventions' (largely relating to stress during and after treatments) being the next highest scoring welfare concern. Due to the open-ended nature of responses in this question section, statistical analysis could not be carried out to relate responses to participant backgrounds. However,

Table 2-1. Top three highest and lowest scoring welfare concerns, depending on participant's production stage-specific experience.

Production stage-specific experience:	Highest scoring welfare concerns:	Lowest scoring welfare concerns:
Freshwater only	Interventions, Handling, Stocking density	Sea lice, Predation, Farm management
Seawater only	Sea lice, Gill health, Environmental challenges	Predation, Interventions, Farm management
Both Freshwater & Seawater	Environmental challenges, Sea lice, Gill health	Farm management, Predation, Stocking density

qualitative differences in weighted scores between welfare concerns were recognised between participants with experience in different production stages (see Table 2-1).

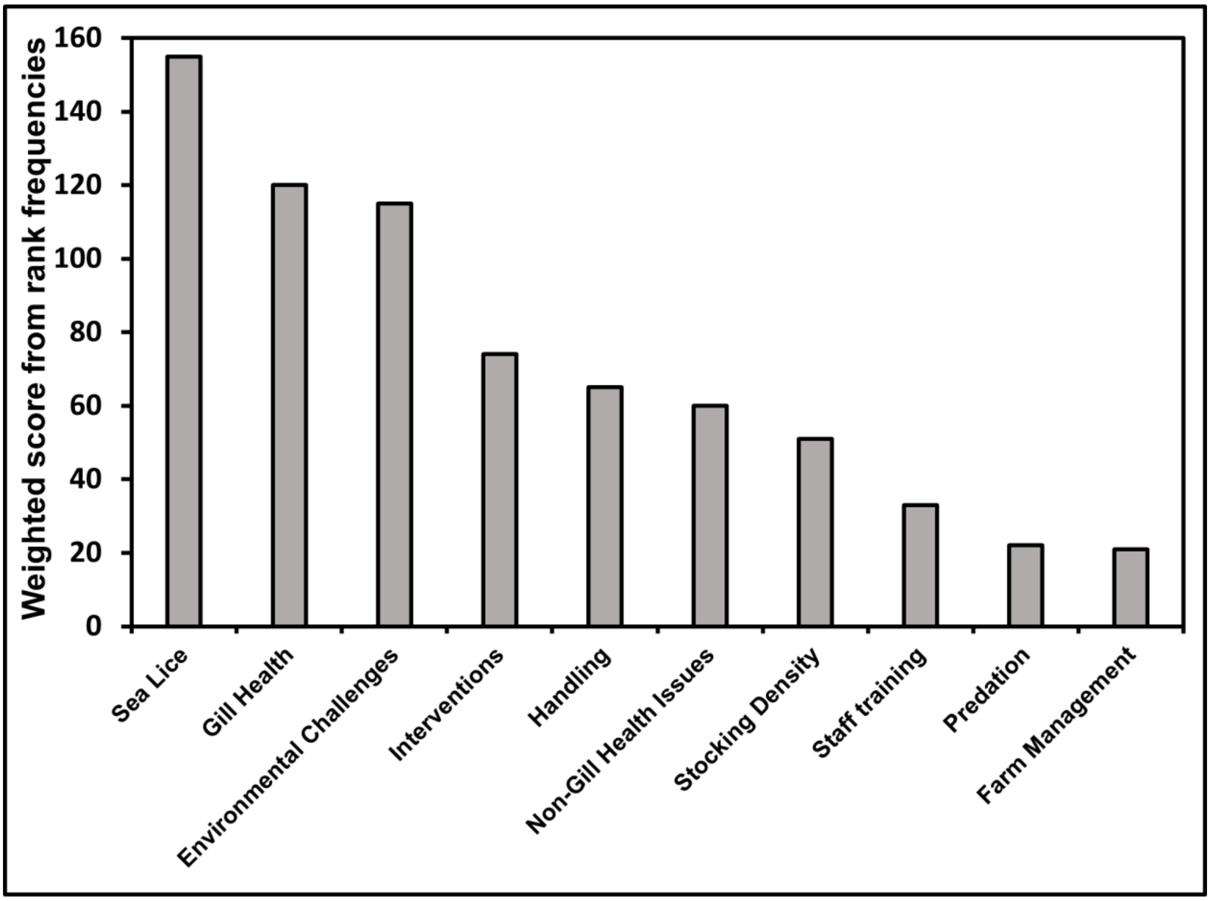

Figure 2-3. Top 10 ranked categories of overall farmed salmon welfare concerns (out of 16 categories listed), based on weighted scores provided by participants (n=61). The open-ended nature of this question meant that statistical differences between categories could not be tested for.

2.4.4 Section 3 – Husbandry practices requiring the most attention

In contrast to welfare concerns, there was far more of an agreement between participants regarding what husbandry practices they considered required the most attention in monitoring salmon welfare. Out of the 12 categories of husbandry practices mentioned by participants, 68% of the total weighted score was accounted for by the top 2 scoring categories (‘Interventions’, and ‘Handling’). The next highest scoring category, ‘Feeding’, accounted for 9% of the total weighted score (see Figure 2-4).

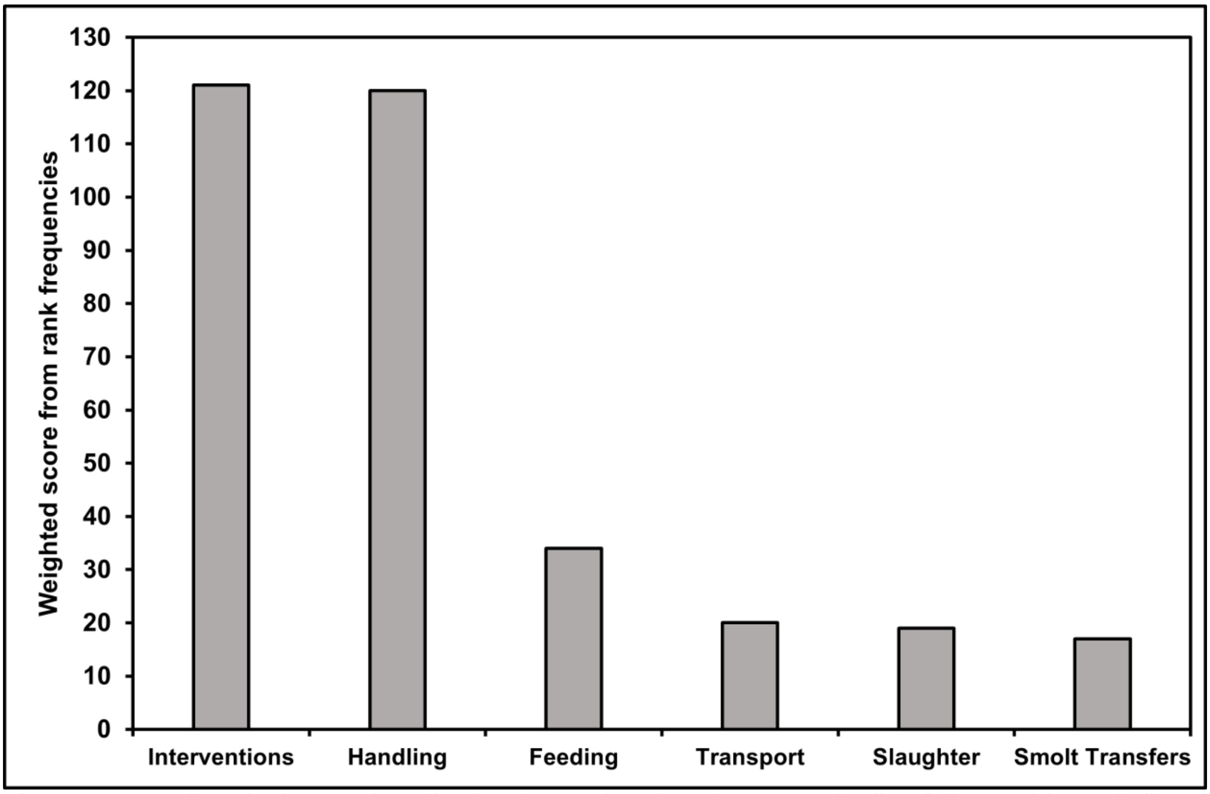

Figure 2-4. Top six ranked categories of husbandry practices (out of 12 categories listed) that participants believed require the most attention in terms of monitoring salmon welfare, based on weighted scores provided (n=61). The open-ended nature of this question meant that statistical differences between categories could not be tested for.

2.4.5 Section 4 – On-farm practicality and effectiveness of welfare measures

Numerically, the 4 highest overall scoring categories of welfare measures were '*Disease/health status of fish by prevalence of conditions during routine observations or sampling of individuals*', '*Changes in behaviour (both routine monitoring and husbandry practices)',* and '*Changes in appetite'.* Significant differences were found between categories in their practicality ratings (Figure 2-5; Kruskal Wallis test: $H = 143.68$, $df = 11$, $P < 0.001$). There was no significant difference found between the 7 highest numerical scoring categories of welfare measures ($P > 0.05$). Three of these categories, however, had the largest number of significant differences found compared to the remaining 9: '*Disease/health status of fish by prevalence of conditions during routine observations or sampling of individuals*', '*Changes in behaviour (routine monitoring)',* and *'Changes in appetite after potentially disturbing husbandry practices'*.

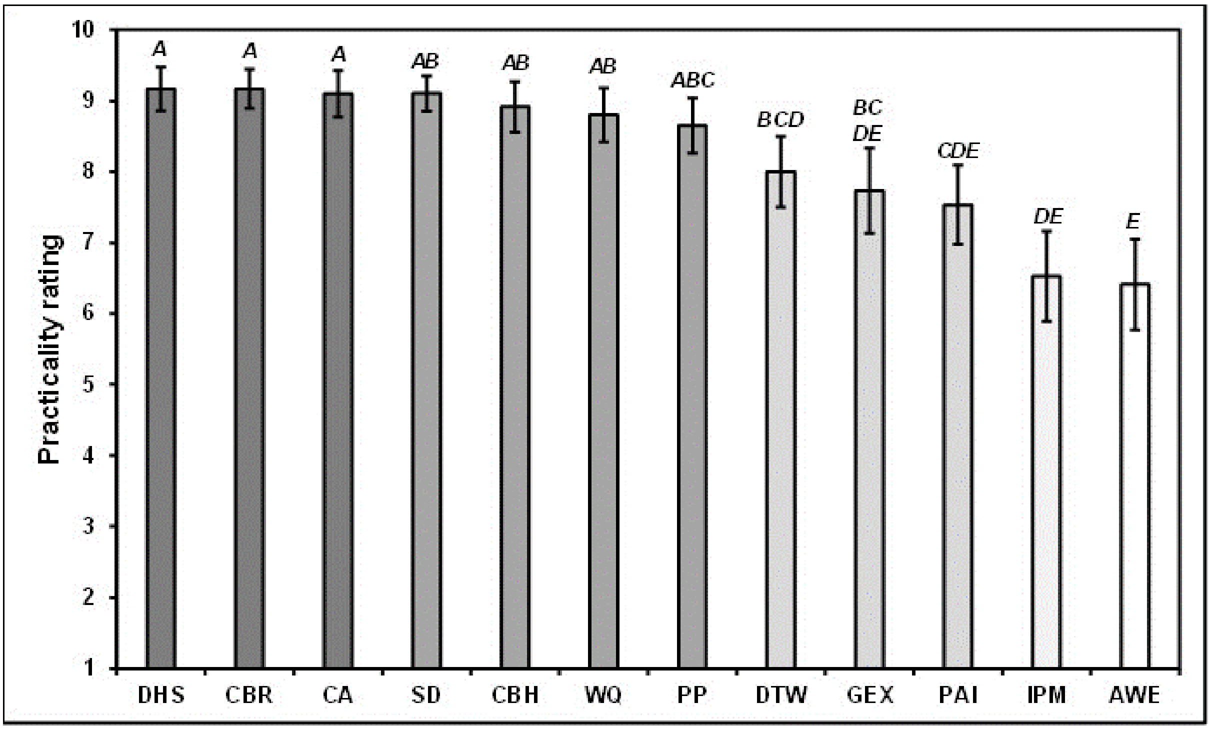

Figure 2-5. Mean practicality ratings of the 12 ranked categories of salmon welfare measures listed, based on ratings provided by participants (n=60). Error bars show 95% confidence intervals. Categories with no matching letters above the error bars indicate a statistical difference (P < 0.05).

DHS	‘Disease/Health status by prevalence of conditions
CBR	‘Changes in behaviour (routine monitoring)’
CA	‘Changes in appetite’
SD	‘Stocking density’
CBH	‘Changes in behaviour (husbandry practices)’
WQ	‘Water quality’
PP	‘Production parameters’
DTW	‘Duration of time out of water’
GEX	‘Grading by external abnormalities’
PAI	‘Presence of acute injuries’
IPM	‘Individual physiological measures of stress’
AWE	‘Assessing welfare by enrichment’

Significant differences were found between categories in their effectiveness ratings (Figure 2-6; Kruskal Wallis test: H = 79.57, df = 11, P < 0.001). There was no significant difference found between the 9 highest scoring categories of welfare measures (P > 0.05). The 3 aforementioned categories, along with ‘*Changes in behaviour (husbandry practices)’* were the highest numerical categories by effectiveness. These 4 categories had the largest number of significant differences found compared to the remaining 8 categories. Pairwise comparisons showed that, for practicality or effectiveness, no single category of welfare measures scored significantly differently from all the other 11 categories.

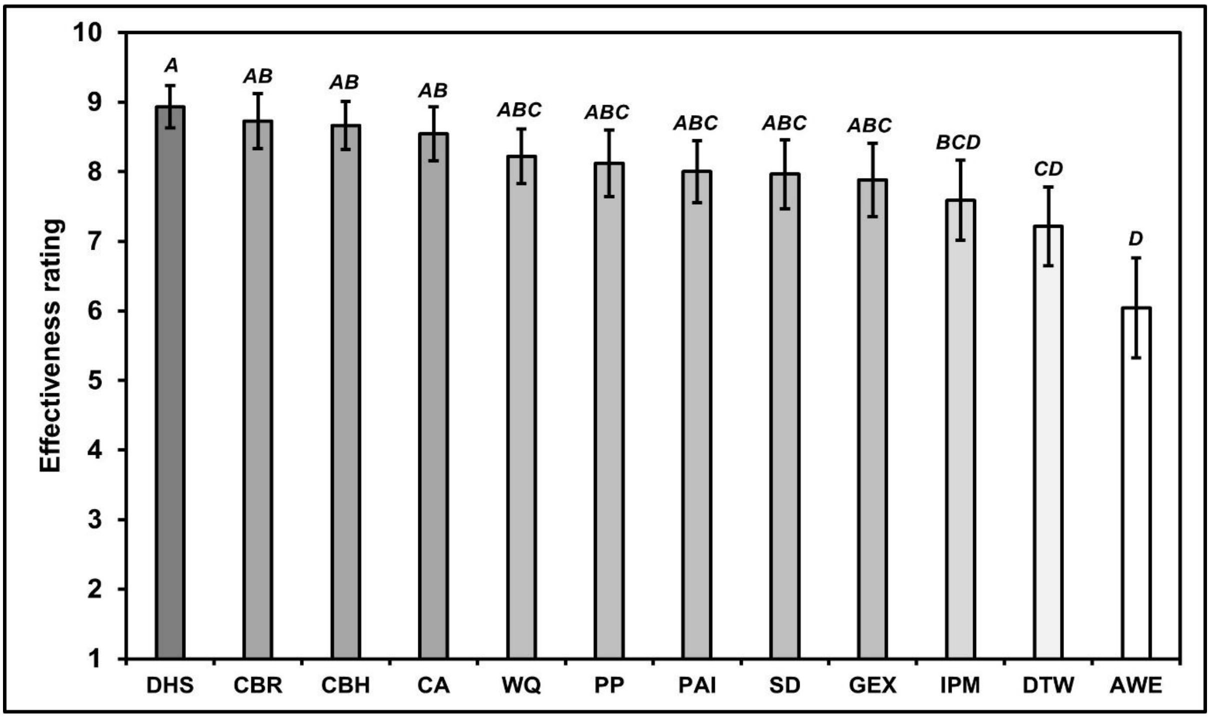

Figure 2-6. Mean effectiveness rating of the 12 categories of salmon welfare measures listed, based on ratings provided by participants (n=59). Error bars show 95% confidence intervals. Categories with no matching letters indicate a statistical difference ($P < 0.05$).

DHS	‘Disease/Health status by prevalence of conditions
CBR	‘Changes in behaviour (routine monitoring)’
CBH	‘Changes in behaviour (husbandry practices)’
CA	‘Changes in appetite’
WQ	‘Water quality’
PP	‘Production parameters’
PAI	‘Presence of acute injuries’
SD	‘Stocking density’
GEX	‘Grading by external abnormalities’
IPM	‘Individual physiological measures of stress’
DTW	‘Duration of time out of water’
AWE	‘Assessing welfare by enrichment’

2.4.5.1 Participants' practical considerations for on-farm use of welfare measures – Thematic analyses

A total of 384 comments were received regarding various considerations about using the listed welfare measures on-site (see Figure 2-7). Comments on how these measures were either ‘*Already taken as part of farming routine’* or ‘*Easy to use and monitor on a consistent basis (if needed)’* accounted for 88 of the 96 statements regarding the ’*Advantages to using welfare measures’.* With the exception of ‘*Assessing welfare by presence/absence of enrichment’*, these comments were made at least once for all other welfare measures listed. Out of the 8 remaining comments regarding advantages, 5 were exclusive to measures involved in ‘*Changes in behaviour’,* stating how such measures could act as early warning signs for arising issues. Conversely, 26 comments were made on ‘*Practicalities regarding use of equipment & facilities’*, all relating to concerns about the necessity for specialist equipment to either facilitate the use of, or even carry out, the listed welfare measures. Of the 188 comments regarding potential ‘*Limitations to using welfare measures effectively’,* 87 stated that ‘*the quality of information depends on the training and motivation of staff involved*’. Such comments were made across all measures, but particularly on those assessing physiological measures of stress, external abnormalities, and changes in behaviour during monitoring and husbandry practices (17, 11, 12, and 10 comments made respectively).

Another 32 comments regarding limitations involved the difficulty of ‘*ensuring a representative sample size*’; these comments were made at least once for all welfare measures that involved assessing the salmon directly. Other limitations mentioned included ‘*inherent subjectivity in the use of the welfare measure’, ‘welfare measure cannot be used in isolation’*, and difficulties in ‘*using the welfare measure to accurately reflect the salmons’ welfare status’*.

There were 53 comments made on the ‘*Practical limitations to using welfare measures on-site*’. Twenty-nine of these stated that certain measures ‘*may require frequent monitoring, which could be costly or time consuming’*. The majority of the 29 comments (17) were specific to assessing physiological measures of stress, external abnormalities, and acute injuries during husbandry practices. Another 22 comments on practical limitations stated that the use of various measures ‘*requires good weather*’. Out of the 21 comments regarding potential ‘*Negative impacts of using welfare measures’*, 15 were made about welfare measures that were likely to require invasive sampling to carry out (assessing physiological measures of stress, external abnormalities, and assessing disease/health status). All 15 of these comments specifically involved concerns about there being a ‘*Significant potential for damage, stress, or mortality to be caused’* to the salmon as a result of using these welfare measures.

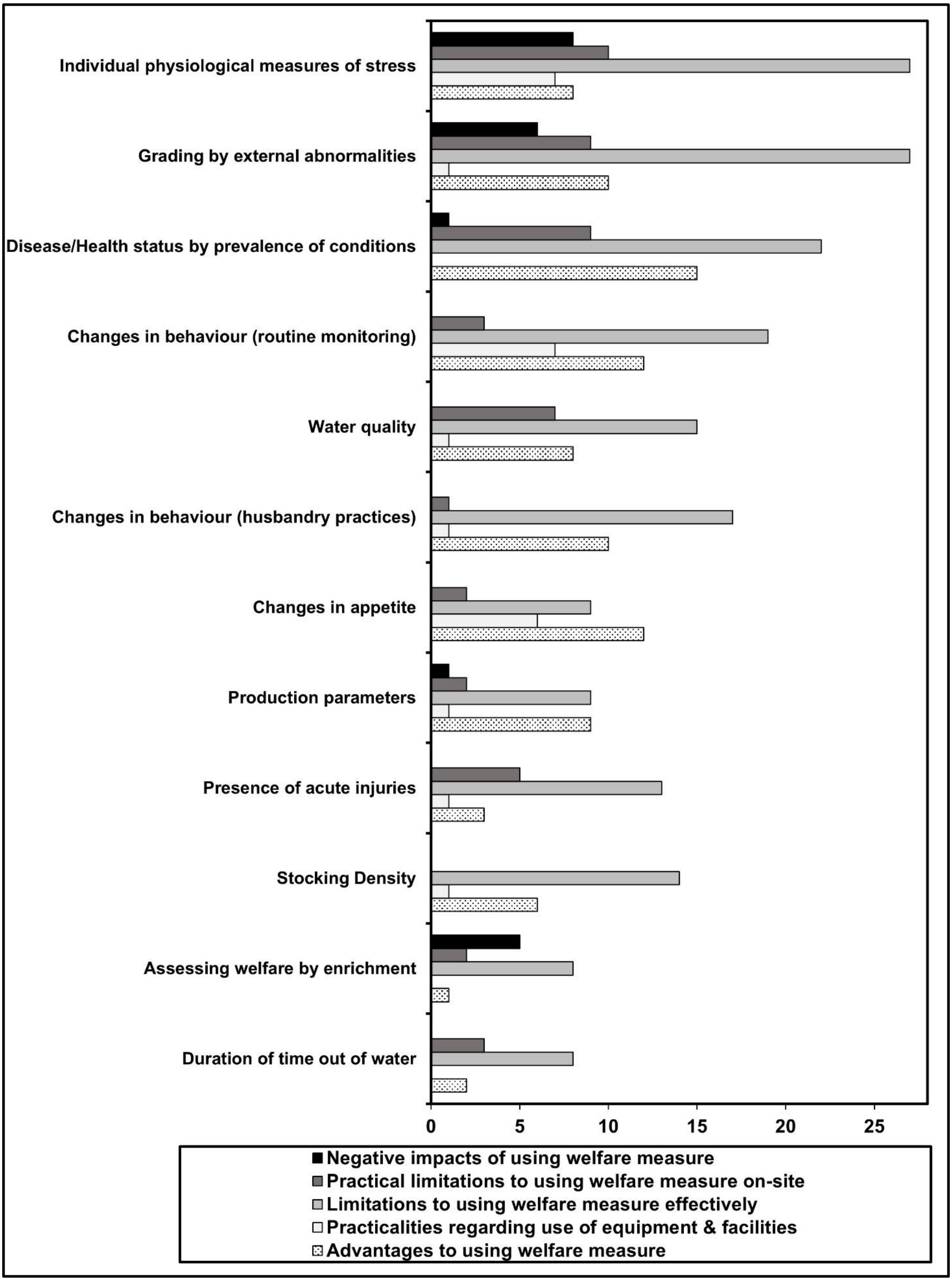

Figure 2-7. Coding frequency for main themes of practicality mentioned by participants (n=53) when given the option for providing comments on the practical considerations of the welfare measures listed.

2.4.6 Section 5 – Relevance and urgency for R&D of welfare assessments

No significant differences were found between the relevance ratings of the different research priorities (Figure 2-8; Kruskal Wallis test: H = 6.56, df = 4, P = 0.161). With regards to urgency ratings, one significant difference was found between the research priority '*Developing welfare indicators that allow for remote monitoring of salmon'* and *'Developing more fish/user friendly methods for welfare indicators which currently require sampling of the fish'* (Figure 2-8; Kruskal Wallis test: H = 13.374, df = 4, P = 0.01.

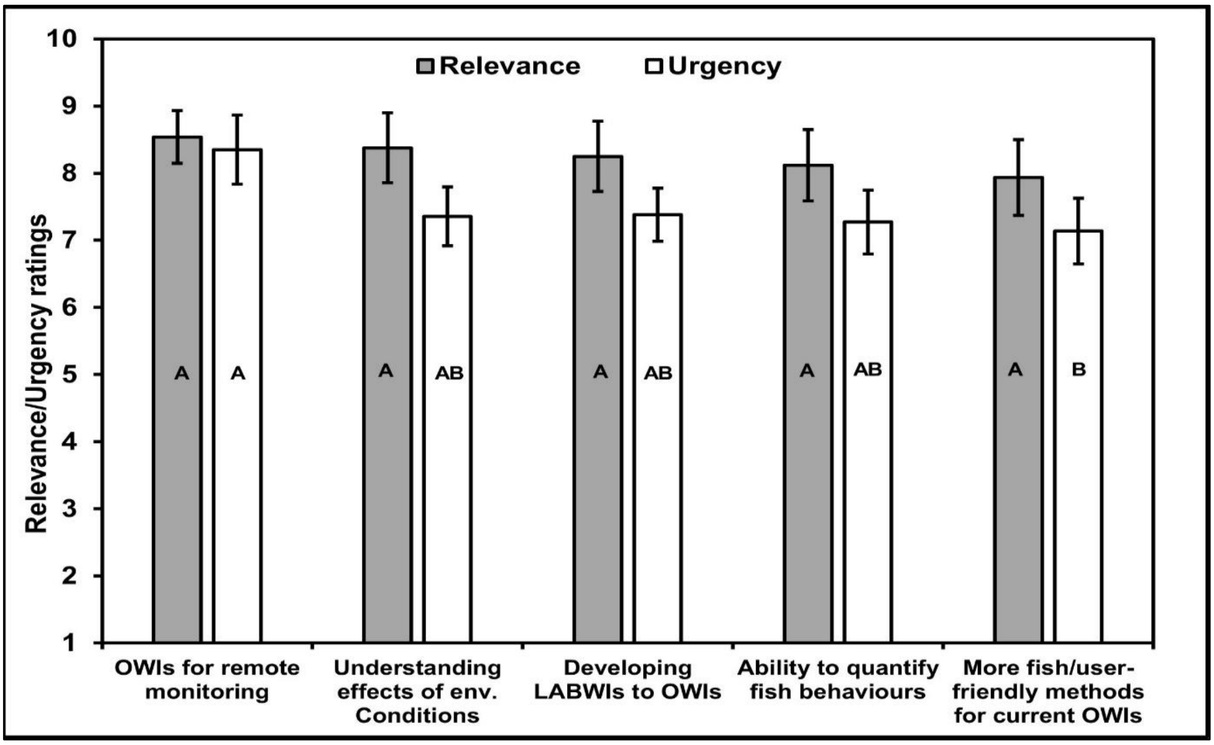

Figure 2-8. Mean relevance and urgency ratings of the five research priorities listed, based on ratings provided by participants (n=59). Error bars show 95% confidence intervals. Research priorities with no matching letters indicate a statistical difference (P < 0.05), with relevance and urgency ratings being compared separately. LABWI = Laboratory-Based Welfare Indicator, OWI = Operational Welfare Indicator.

2.4.7 Section 6 – Farming routines most practical for monitoring salmon welfare

Out of all routines, 'Health checks' and 'Feeding times' accounted for 61% of the total routines mentioned as being the most practical as an opportunity to assess welfare (see figure 2-9). In comparison, 'Routine cage/tank inspection', 'Video monitoring', and 'Grading and/or transfer' collectively accounted for 36% of the routines selected. Any mentions of routines by participants outside of the list provided ('Other') accounted for just 4% of total routines selected. Any mentions of routines by participants outside of the list provided (‘Other’) accounted for just 4% of total routines selected.

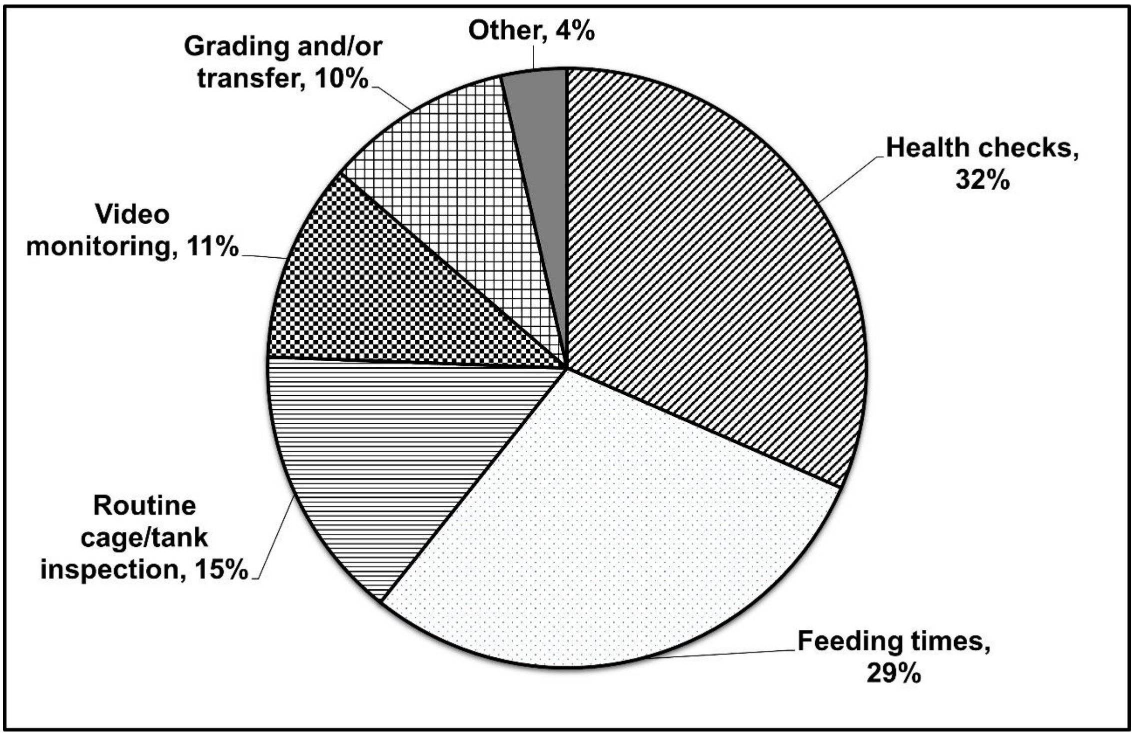

Figure 2-9. Most suitable opportunities for monitoring welfare measures on site during a farm’s daily routine, based on the relative proportion of times they have been mentioned by participants (n=60) as a suitable opportunity for monitoring certain welfare measures on-site.

2.5 Discussion

Ascertaining what best approaches there are to the assessment and management of on-farm salmon welfare issues is, ultimately, one of the first steps towards addressing these issues. The results from this survey represent opinions on this matter from professionals across various production stages within the Scottish salmon farming industry. Owing to the sample size, this survey cannot claim to be representative of the industry’s views as a whole. However, the variety of farming experience of the participants involved is an encouraging sign that the survey has succeeded in obtaining valuable insights from a diverse range of professionals directly involved with farmed salmon. Despite such variation in experience between participants, and some differences on what constitutes the largest overall concerns facing farmed salmon, there was a strong consensus on what areas of welfare monitoring and research priorities the industry must focus on to safeguard the future of farmed salmon welfare.

When participation in a survey is voluntary, it is important to reduce recruitment bias wherever possible (Fox, Hunn and Mathers, 2009). From the combination of the various recruitment methods, particularly with some of the largest salmon producers in Scotland agreeing to contact their entire production team to encourage participation, a reliable assumption can be made that as many of the Scottish salmon production staff as possible were at least informed of the opportunity to participate. In terms of reducing any systematic bias introduced by those individuals who chose to participate, the variety of professional backgrounds involved in the survey also suggests that this bias was limited.

2.5.1 Key areas of concern within salmon farming

Seawater rearing received one of the highest scores of relative importance, and this was not solely explained for by the largest proportion of respondents having seawater experience. Participants with only freshwater experience still scored the seawater rearing and freshwater stage almost identically (<1% difference in total weighted score). The relative importance of salmon welfare during seawater rearing may be partly due to this stage representing the largest portion of the salmon's overall life cycle (Superior Fresh, 2019; Scottish Sea Farms, 2021). There are also key welfare concerns specific to this stage which may further explain the participants' views on its importance. Sea lice, which received the highest numerical weighted score of welfare concerns in this survey, are also present exclusively in this production stage. Sea lice have a longstanding reputation as one of the largest welfare risks to farmed salmon in the marine environment, and as one of the most damaging parasites to the salmonid farming industry worldwide (Costello, 2006; Brown *et al.*, 2008). Infestations are known to cause physical damage to the host's skin, potentially leading to reduced appetite and growth, as well as increased physiological stress through osmoregulatory dysfunction (Thorstad *et al.*, 2015; Abolofia, Asche and Wilen, 2017). A further indirect consequence of sea lice are delousing operations, particularly through mechanical and thermal methods, which have been known to impact salmon welfare and, in some cases, lead to increased mortality rates (Overton *et al.*, 2018, 2019). This concern was also reflected by 9 of the participants in this survey.

Regardless of how important participants believe that the seawater rearing stage is for overall salmon welfare, it is important to recognise that each production stage listed still scored relatively highly in terms of importance by each cohort of participants. Therefore, similar consideration must still be given to salmon welfare during all production stages.

Gill health was the second largest concern for welfare, concurring with the growing concern over poor welfare and increasing losses related to gill disease in Atlantic salmon worldwide (Mitchell and Rodger, 2011; Gjessing *et al.*, 2017). A monthly mortality report by the SSPO in June (SSPO, 2021) showed that where a Scottish farm listed a mortality rate of 3.4% or higher, it was linked to either gill health, gill management (e.g., treatments for gill health) or viral challenges. The three highest mortality rates listed (9.5%, 7.2%, and 5.7%) were all related to gill health issues. Gills are naturally exposed to the constantly changing physico-chemical properties of the surrounding water, as well as to numerous aetiological agents such as algal blooms, jellyfish swarms, viruses, and bacteria that can compromise gill health (Steinum *et al.*, 2010; Baxter *et al.*, 2011; Mitchell and Rodger, 2011; Rodger, Henry and Mitchell, 2011; Gjessing *et al.*, 2017). 'Complex gill disease' has also become a growing issue for farmed salmon, particularly in the marine environment over the past few years (Herrero *et al.*, 2018; Boerlage *et al.*, 2020).

The degree of concern relating to environmental challenges was comparable to that of gill health. In a welfare risk assessment carried out for EFSA, abiotic hazards (mainly water quality) were a concern across all life stages of Atlantic salmon (Brown *et al.*, 2008). Welfare concerns relating to environmental challenges in both this survey and the EFSA risk assessment mostly included concerns about water quality, as well as the issue of ensuring that appropriate enclosures were used and maintained. This includes selecting suitable site locations for sea cages. With sea cages being exposed to uncontrollable environments, water currents and low water O_2 content have previously been identified as the abiotic hazards with the most potential to affect the physiology, behaviour, and ultimately welfare of farmed salmon (Brown *et al.*, 2008; Hvas, Folkedal and Oppedal, 2021).

2.5.1.1 Freshwater production staff highlighted the importance of interventions & handling

Responses regarding welfare concerns were the most varied in this survey when compared against participants' experience in specific production stages. Considering that the survey had significantly more participants with seawater experience, the overall scores for welfare concerns may have represented concerns that can be found more within seawater rearing. Therefore, concerns listed by participants with only freshwater experience have been considered separately.

In contrast to other participants, freshwater production staff ranked sea lice as one of the three lowest concerns for salmon welfare. Since sea lice exclusively affect the seawater stage, staff lacking first-hand experience in dealing with this parasite may not appreciate the true extent of their impacts. Environmental challenges were also far less of a concern to freshwater staff, potentially due to environmental parameters being easier to control in freshwater systems compared to seawater cages (Brown *et al.*, 2008). Instead, interventions (largely relating to treatments) of salmon were their highest overall welfare concern, followed by handling and stocking density. The immediate impacts from invasive events such as treatments, vaccinations, and handling may be more visible to freshwater production staff, and could potentially explain why they ranked these welfare concerns much higher.

The importance of interventions and handling was also reflected in which husbandry practices participants believed required the most attention in terms of monitoring salmon welfare. Across all groups of participants, interventions and handling were of the highest priority. Various handling procedures can lead to acute stress, injury, weakened osmoregulatory abilities, and increased disease incidence in salmon (Ashley, 2007; Brown *et al.*, 2008; Powell, Reynolds and Kristensen, 2015). Fish suffering from disease or injury are already under physiological stress, and are therefore susceptible to the cumulative stress that can occur during certain treatments (Marcos-López *et al.*, 2017)**.** Careful monitoring of salmon welfare is therefore required during interventions and any handling prior to these practices must be minimised due to the high risk of impact to health and welfare at these times.

2.5.1.2 Discrepancies in perceived importance of husbandry practices and concerns; staff knowledge, staff training, slaughter, and transport

The importance of staff training and biosecurity for salmon welfare across all life stages has been frequently mentioned in previous studies (Brown *et al.*, 2008). Through interviews of employees at various company levels, Størkersen et al. (2021) concluded that daily tasks on-site were considered to make the most positive contribution to fish welfare. Production staff play an important role by sharing knowledge, developing, and executing routines to protect farmed fish (Størkersen *et al.*, 2021). However, participants in this survey were more concerned with the issues mentioned above (sea lice, gill health, environmental challenges, risks associated with interventions) than with staff training and farm management. This discrepancy may partially be the result of participants being limited to listing only 3-5 of their most significant welfare concerns *facing* salmon welfare. Rather than dismissing the importance of training and management, these may have simply been less important to the participants than animal-based concerns that directly affect the salmon. In addition, handling and environmental challenges potentially overlap with concerns relating to staff training and farm management, which could further explain their underrepresentation in these results.

Overall, participants in this survey also scored transport and slaughter far lower than interventions, handling, or even feeding. This is in stark contrast to the literature, which have often considered processes relating to slaughter and transport as serious threats to welfare (Poli *et al.*, 2005; Erikson *et al.*, 2016). Participants in this survey may have treated any of the handling, crowding, or grading that occurs prior to these two practices as separate to the actual slaughter/transport process themselves. The procedures immediately prior to slaughter/transport could potentially account for a large portion of the concern associated with them. This difference in opinion may also be partially explained by the fact that transport and slaughter represent a relatively small fraction of the salmon’s overall life cycle. In comparison, examples of interventions or handling can occur many times over, leading to a larger cumulative effect on the salmon’s overall welfare status.

Variation was also found in welfare concerns between participants with different farming experience, and this in turn may be related to the specific challenges faced in each stage of production (Noble et al., 2018). When concerns vary between stakeholders and even within the industry, identifying welfare priorities becomes complex. Although certain welfare concerns have been identified in this survey as the “largest” concerns by participants (e.g., sea lice, gill health, environmental challenges, risks associated with interventions), this serves mainly to inform on some of the major concerns present in Scottish salmon farming. At the very least, equal consideration must still be given to any of the welfare concerns from each production stage and husbandry practice for which participants have repeatedly mentioned.

2.5.1.3 Categorisation of open-ended responses

In order to examine open-ended responses on welfare concerns and husbandry practices requiring the most attention, participants’ responses were categorised by their degree of similarity to each other. Any inherent subjectivity behind how these groupings were made ran the risk of certain categories being misrepresented by their total weighted score. The total weighted score attributed to a specific concern of husbandry practice could be over / underrepresented, depending on how broad / narrow of a category the responses were grouped under. The broader a topic involved within a category (e.g., environmental challenges), the larger the variety of responses that could potentially be included, thus inflating the weighted score in relation to other categories. Extra care was therefore taken to ensure that the categories only include responses that were as close to as identical as possible to minimise what impact this could have. Statistical analyses could not be carried out on these open-ended responses to determine whether they were significantly correlated to participants’ backgrounds. Outside of the large contrasts in welfare concerns between different cohorts, however, the high degree of consensus found between participants suggests that any potential influences on responses were less of a concern.

2.5.2 Welfare monitoring and assessment – key areas of focus

2.5.2.1 Suitability of on-farm welfare measures for non-invasive, remote monitoring

Participant responses indicated that, for the majority of welfare measures, there was no difference between their practicality or effectiveness. No single category was statistically different from all remaining 11 categories in either rating. However, welfare measures relating to monitoring changes in behaviour, appetite, or the disease/health status of the salmon were found within the highest scoring group of categories. Collectively, these categories of welfare measures had significant differences with the largest number of other categories in both practicality and effectiveness ratings. Out of the categories listed, these welfare measures also constitute a broader class of animal-based, non-invasive measures that can be monitored remotely.

While the ratings produced some quantifiable indication of how appropriate these measures are for on-farm use, additional comments gave participants' the opportunity to give further detail on this topic. With the exception to assessing welfare by the presence/absence of enrichment, all remaining measures listed were mentioned at least once as having the advantage of either already being recorded on-site or able to be readily measured as part of the farming routine. This is reflected in the high practicality scores across the majority of measures listed. The group of animal-based, non-invasive measures that can be monitored remotely continued to maintain a more positive sentiment around their use on-site. More than half of the remaining comments regarding advantages to using these welfare measures were exclusive to monitoring changes in behaviour. Participants also believed that the use of these measures posed fewer risks for salmon welfare compared with other animal-based measures. This is in accordance with the previously mentioned sentiment (in 2.5.1.1) that handling of the salmon must be minimised. Additionally, monitoring changes in behaviour may also provide early warning signs for issues that arise on-site (Huntingford *et al.*, 2006; Oppedal, Dempster and Stien et la., 2011). When compared with other direct animal-based measures of salmon welfare, the frequent monitoring that may be required for non-invasive measures (monitoring changes in behaviour or appetite) were seen as not being as costly or time-consuming.

Participants' responses suggest that welfare measures that involve handling or invasive procedures of the salmon (e.g., sampling individuals for physiological measures of stress) should be limited, unless they are an essential part of the production process. Regular health checks are now regarded as a crucial aspect of farming routines for protecting health and welfare for salmon (Rey Planellas, Little and Ellis, 2019; RSPCA, 2021). This likely explains why participants deemed health checks as one of the most suitable opportunities for monitoring welfare, due to the valuable welfare-relevant information they already provide. As health checks are already required, they provide an opportunity to use valuable animal-based measures (e.g., fin damage, sea lice infestation, body/skin condition) without causing unnecessary stress. For all animal-based welfare measures, however, participants noted a number of limitations. Any measures involving a direct assessment of the salmon face the challenge of obtaining a representative sample of the fish. Specialist equipment may also often be required. The most frequently mentioned limitation when using these animal-based measures was their dependency on the motivation and training of staff. This is in contrast to the low ratings that staff knowledge and training received as an overall welfare concern. This suggests that, while participants appreciated the importance of staff training and knowledge relating to monitoring and safeguarding salmon welfare, they did not believe that this was currently a major concern to farmed salmon welfare. Participants also recognised the importance of using multiple measures to avoid the subjective bias that may arise from any single measure (Sneddon, Braithwaite and Gentle, 2003b).

Practicality and effectiveness ratings did not provide any information on the need for further developments. In order to identify areas of welfare assessment that are both appropriate for on-farm use, and require further development, these ratings have to be considered with the identified research priorities.

2.5.2.2 Key areas of development in welfare monitoring and assessment

All research priorities were deemed equally relevant for improving the monitoring and safeguarding of salmon welfare. Given their equal relevance, they can only be differentiated by their urgency ratings. The development of remote monitoring was seen as the most urgent, which may have been highlighted to participants by the restricted access to sites for farm staff during the 2020 COVID-19 pandemic (Murray *et al.*, 2021). These restrictions would have likely had a significant impact on the degree of active surveillance that was possible during the lockdown period, with in-person audits being replaced with virtual assessments for 2 months (FishFarmingExpert, 2020; Murray *et al.*, 2021). Relying on virtual assessments could hinder the ability for certification bodies to safeguard salmon welfare due to the limited amount of information that can be obtained. These events have likely demonstrated the necessity of having welfare measures that can be used without requiring staff on-site. This would include passive, non-invasive measures that could be recorded through the use of remote sensors, or video and acoustic monitoring (Føre, Alfredsen and Gronningsater, 2011; Brijs *et al.*, 2021; Bell *et al.*, 2022). High urgency ratings for remote monitoring as a research priority suggest that measures currently available may not yet be developed enough to fulfil this role.

2.5.2.3 Improving non-invasive, animal-based and remote welfare monitoring on-site: a case for behavioural welfare measures

Behavioural measures were identified as a promising candidate for non-invasive and remote welfare monitoring. The potential benefits of their implementation into practical farm-management strategies have already been acknowledged (Dawkins, 2003; Huntingford *et* *al.*, 2006; Oppedal, Dempster and Stien, 2011; Martins *et al.*, 2012; Miller *et al.*, 2020; Barreto *et al.*, 2021; O'Donncha *et al.*, 2021). Although direct measures of animal welfare tend to be the most informative, their use often comes with the cost of either being time-consuming, technically complex, or causing disturbances to the fish (Huntingford *et al.*, 2006). In contrast, behavioural indicators are one of the few animal-based measures that benefit from being comparatively fast and easy to observe (Huntingford *et al.*, 2006; Martins *et al.*, 2012). Effective inclusion of behavioural indicators with other evidence of an animal's health could help to identify pre-clinical signs of health problems (Dawkins, 2003). Improving the ability for farm staff to recognise and prevent problems before they can severely impact stock is beneficial not only to the fish, but for farm production. Further innovations in camera technology and image processing may allow for significantly improved on-farm surveillance of salmon behaviour (Saberioon *et al.*, 2017).

While video monitoring accounted for just 11% of the routines mentioned as most suitable for monitoring salmon welfare, it is important to consider that camera systems are already routinely used to monitor feeding and swimming behaviours in commercial aquaculture facilities (Pinkiewicz, Purser and Williams, 2011). Feeding times, which accounted for 29% of the routines mentioned, also provide opportunities for assessing behavioural patterns either through video or acoustic devices (Martins *et al.*, 2012; Hassan *et al.*, 2019). It is not clear if scientific research could ever provide a robust measure of salmon's subjective experiences (Mason and Mendl, 1993; Fraser *et al.*, 1997; Broom, 1998; Dawkins, 1998; Jarvis *et al.*, 2021). Behavioural analysis is currently the only tool which provides any relevant insights (Turnbull and Kadri, 2007; Folkedal *et al.*, 2012; Martins *et al.*, 2012; Zhao, Bao, Zhang, Zhu, Liu, Lu, *et al.*, 2018; Hassan *et al.*, 2019). A promising approach for gaining such insights is Qualitative Behavioural Assessment (QBA), which describes and quantifies expressive qualities of an animal's dynamic body language using qualitative behavioural terms (Jarvis *et al.*, 2021). There are, however, risks of misinterpreting changes in behaviours (Weary and Fraser, 1995; Dawkins, 2003). Welfare assessments should therefore not rely solely on behaviour or any single welfare measure, and rather use an integrated approach of various measures (Jarvis *et al.*, 2021).

2.6 Conclusion

In terms of key areas of focus for salmon welfare, seawater rearing and sea lice seem to be of particular importance. Gill health and environmental challenges (mainly relating to water quality) are two other key welfare concerns perceived to threaten salmon welfare. Participants emphasised the importance of monitoring salmon welfare during husbandry practices where contact and disturbance to the fish is unavoidable, particularly during handling and interventions. Further reflecting the importance of minimised handling, this survey has identified that non-invasive, animal-based welfare measures (particularly those involving behavioural assessment) as one of the most opportune areas for further developing the practicality and efficacy of on-farm salmon welfare assessments.

The results from this survey have also exemplified that no single measure allows for a comprehensive assessment of farmed salmon welfare, and that there are significant welfare concerns which can be unique to a husbandry stage or practice. Protecting farmed salmon welfare will therefore depend on the industry's ability to address the major concerns specific to each of these. This reflects the importance of using an integrated approach to welfare assessments that combines behavioural, physiological, and production-based parameters. Future research should examine potential relationships between behavioural and physiological welfare measures to help validate the use of behavioural assessments when interpreting the welfare status of salmon.

The economic and social aspects of any industry are well established dimensions of its sustainability (UN General Assembly, 2015). With regards to the Scottish salmon farming industry, the public's perception of welfare issues are central to both of these pillars. This survey has helped provide direction for further developing the practicality and efficacy of on-farm welfare assessment and management, and has therefore contributed one step further to advancing farmed salmon welfare. As a result of aiding social acceptance through improved salmon welfare, this work will further add to the potential sustainability of salmon aquaculture.

2.7 Acknowledgements

We gratefully acknowledge all volunteers from the Scottish salmon farming industry who took their time to provide their opinions and insights for this project. This research was funded by the University of Stirling, University of Edinburgh, and SRUC.

2.8 Appendix

Supplementary data to this article can be found at http://hdl.handle.net/11667/201. Additional copy of survey embedded in the final section of this thesis.

2.9 References

Abolofia, J., Asche, F. and Wilen, J. E. (2017) The cost of lice: Quantifying the impacts of parasitic sea lice on farmed Salmon. Marine Resource Economics, 32(3). doi: https://doi.org/10.1086/691981.

Animal Welfare Act 2006 (2006) Animal Welfare Act 2006, The National Archives. Available at: https://www.legislation.gov.uk/ukpga/2006/45/contents (Accessed: 27 June 2022).

Ashley, P. J. (2007) Fish welfare: Current issues in aquaculture. Applied Animal Behaviour Science, 104(3–4), pp. 199–235. doi: 10.1016/j.applanim.2006.09.001.

Barreto, M. O. et al. (2021) Emerging indicators of fish welfare in aquaculture. Reviews in Aquaculture, pp. 1–19. doi: 10.1111/raq.12601.

Baxter, E. J. et al. (2011) Gill damage to Atlantic Salmon (Salmo salar) caused by the common jellyfish (Aurelia aurita) under experimental challenge. PLoS ONE, 6(4), pp. 4–9. doi: 10.1371/journal.pone.0018529.

Bell, J. L. et al. (2022) Environmental monitoring tools and strategies in salmon net-pen aquaculture. Integrated Environmental Assessment and Management, 18(4), pp. 950–963. doi: 10.1002/ieam.4622.

Bergqvist, J. and Gunnarsson, S. (2013) Finfish Aquaculture: Animal Welfare, the Environment, and Ethical Implications. Journal of Agricultural and Environmental Ethics, 26(1), pp. 75–99. doi: 10.1007/s10806-011-9346-y.

Boerlage, A. S. et al. (2020) Epidemiology of marine gill diseases in Atlantic salmon (Salmo salar) aquaculture: a review. Reviews in Aquaculture, 12(4), pp. 2140–2159. doi: 10.1111/raq.12426.

Brijs, J. et al. (2021) Bio-sensing technologies in aquaculture: how remote monitoring can bring us closer to our farm animals. Philosophical Transactions of the Royal Society B, 376(1830). doi: https://doi.org/10.1098/rstb.2020.0218.

Broom, D. M. (1998) Welfare, stress, and the evolution of feelings. Advances in the Study of Behavior, pp. 317–403.

Broom, D. M. (2011) A History of Animal Welfare Science. Acta Biotheoretica, 59(2), pp. 121–137. doi: 10.1007/s10441-011-9123-3.

Brown, D. M. et al. (2008) Animal welfare aspects of husbandry systems for farmed Atlantic salmon - Scientific Opinion of the Panel on Animal Health and Welfare. The EFSA Journal. doi: 10.2903/j.efsa.2008.796.

Costello, M. J. (2006) Ecology of sea lice parasitic on farmed and wild fish. Trends in Parasitology, 22, pp. 475–483. doi: 10.1016/j.pt.2006.08.006.

Dawkins, M. S. (1998) Evolution and animal welfare. The quarterly review of biology, 73(3), pp. 305–328.

Dawkins, M. S. (2003) Behaviour as a tool in the assessment of animal welfare. Zoology, 106(4), pp. 383–387. doi: 10.1078/0944-2006-00122.

Dawkins, M. S. (2004) Using behaviour to assess animal welfare. Animal Welfare, 13(SUPPL.), pp. 3–7.

Dawkins, M. S. (2006) A user's guide to animal welfare science. Trends in Ecology and Evolution, 21(2), pp. 77–82. doi: 10.1016/j.tree.2005.10.017.

Economics, B. (2020) Aquaculture sector - wider economic impacts: estimates. Available at: https://www.gov.scot/publications/estimation-wider-economic-impacts-aquaculture-sector-scotland/ (Accesed 20th October 2021).

Ellis, T. et al. (2016) Trends during development of Scottish salmon farming: An example of sustainable intensification?. Aquaculture, 458, pp. 82–99. doi: 10.1016/j.aquaculture.2016.02.012.

Erikson, U. et al. (2016) Crowding of Atlantic salmon in net-pen before slaughter. Aquaculture, 465, pp. 395–400. doi: 10.1016/j.aquaculture.2016.09.018.

FAWC (2014) Opinion on the Welfare of Farmed Fish. FAWC report, pp. 1-40. Available at: https://assets.publishing.service.gov.uk/media/5a7eb32340f0b62305b829c4/Opinion_on_the_welfare_of_farmed_fish_at_the_time_of_killing.pdf (Accessed: 11 October 2021).

Fife-Cook, I. and Franks, B. (2019) Positive Welfare for Fishes: Rationale and Areas for Future Study. Fishes, 4(2), p. 31. doi: 10.3390/fishes4020031.

Fisher, M. W. (2009) Defining animal welfare — does consistency matter?. New Zealand Veterinary Journal, 57(2), pp. 71–73. doi: 10.1080/00480169.2009.36880.

FishFarmingExpert (2020) RSPCA Assured targets return of farm visits, FishFarmingExpert News. Available at: https://www.fishfarmingexpert.com/article/rspca-assured-takes-first-step-towards-normal-assessments/ (Accessed: 11 October 2021).

Folkedal, O., Stien, L. H., Torgersem, T. et al. (2012) Food anticipatory behaviour as an indicator of stress response and recovery in Atlantic salmon post-smolt after exposure to acute temperature fluctuation. Physiology and Behavior, 105(2), pp. 350–356. doi: 10.1016/j.physbeh.2011.08.008.

Føre, M., Alfredsen, J. A. and Gronningsater, A. (2011) Development of two telemetry-based systems for monitoring the feeding behaviour of Atlantic salmon (Salmo salar L.) in aquaculture sea-cages. Computers and Electronics in Agriculture, 76(2), pp. 240–251. doi: 10.1016/j.compag.2011.02.003.

Fox, N., Hunn, A. and Mathers, N. (2009) Sampling and Sample Size Calculation. The NIHR RDS for the East Midlands / Yorkshire & the Humber, 1(1), pp. 1–41. Available at: https://www.gov.scot/publications/scottish-fish-farm-production-survey-2019/pages/5/. (Accessed: 30 October 2021).

Fraser, D. et al. (1997) A scientific conception of animal welfare that reflects ethical concerns. Animal Welfare, 6(3), pp. 1–19.

Gjessing, M. C. et al. (2017) Salmon gill poxvirus, a recently characterized infectious agent of multifactorial gill disease in freshwater- and seawater-reared Atlantic salmon. Journal of Fish Diseases, 40(10), pp. 1253–1265. doi: 10.1111/jfd.12608.

Hassan, W. et al. (2019) Internet of Fish: Integration of acoustic telemetry with LPWAN for efficient real-time monitoring of fish in marine farms. Computers and Electronics in Agriculture, 163, pp. 1–9. doi: 10.1016/j.compag.2019.06.005.

Herrero, A. et al. (2018) Complex gill disease: an emerging syndrome in farmed Atlantic salmon (Salmo salar L.). Journal of Comparative Pathology, 163, pp. 23–28. doi: 10.1016/j.jcpa.2018.07.004.

Huntingford, F. A. et al. (2006) Current issues in fish welfare. Journal of Fish Biology, 68, pp. 332–372. doi: 10.1111/j.1095-8649.2005.01046.x.

Hvas, M., Folkedal, O. and Oppedal, F. (2021) Fish welfare in offshore salmon aquaculture. Reviews in Aquaculture, 13(2), pp. 836–852. doi: 10.1111/raq.12501.

Jarvis, S. et al. (2021) Qualitative behavioral assessment in juvenile farmed Atlantic salmon (Salmo salar): Potential for On-Farm Welfare Assessment. Frontiers in Veterinary Science, 8, pp. 1–11. doi: 10.3389/fvets.2021.702783.

Kenyon, W. and Davies, D. (2018) Salmon Farming in Scotland. Available at: https://digitalpublications.parliament.scot/ResearchBriefings/Report/2018/2/13/Salmon-Farming-in-Scotland (Accessed: 30 October 2021).

Lafferty, K. D. et al. (2015) Infectious Diseases Affect Marine Fisheries and Aquaculture

Economics. Annual Review of Marine Science, 7, pp. 471–496. doi: https://doi.org/10.1146/annurev-marine-010814-015646.

Manfrin, A., Messori, S. and Arcangel, G. (2018) Strengthening fish welfare research through a gap analysis study. Report for SCAR FISH and SCAR CWG. Animal Health and Welfare Research.

Marcos-López, M. et al. (2017) Oxidative stress is associated with late-stage amoebic gill disease in farmed Atlantic salmon (Salmo salar L.). Journal of Fish Diseases, pp. 1–6. doi: 10.1111/jfd.12699.

Marschall, E. A. et al. (1998) A framework for understanding Atlantic salmon (Salmo salar) life history. Canadian Journal of Fisheries and Aquatic Sciences, 55(SUPPL.1), pp. 48–58. doi: 10.1139/cjfas-55-s1-48.

Marsden, P. V. and Wright, J. D. (2010) Handbook of Survey Research. 2nd edn. Emerald Group Publishing.

Martins, C. I. M. et al. (2012) Behavioural indicators of welfare in farmed fish. Fish Physiology and Biochemistry, 38(1), pp. 17–41. doi: 10.1007/s10695-011-9518-8.

Mason, G. and Mendl, M. (1993) Why is there no simple way of measuring animal welfare?. Animal welfare, 2(4), pp. 301–319.

Mellor, D. J. (2016) Updating animal welfare thinking: Moving beyond the “Five Freedoms” towards “A Life Worth Living”. Animals, 6(3), pp. 1–20. doi: 10.3390/ani6030021.

Mellor, D. J. and Beausoleil, N. J. (2015) Extending the “Five Domains” model for animal welfare assessment to incorporate positive welfare states. Animal Welfare, 24(3), pp. 241–253. doi: 10.7120/09627286.24.3.241.

Mellor, D. J. and Reid, C. S. W. (1994) Concepts of animal well-being and predicting the impact of procedures on experimental animals. Improving the well-being of animals in the research environment, pp. 3–18. Available at: http://org.uib.no/dyreavd/harm-benefit/Concepts of animal well-being and predicting.pdf. (Accessed: 20th October, 2021).

Miller, L. J. et al. (2020) Behavioral diversity as a potential indicator of positive animal welfare. Animals, 10(7), pp. 1–17. doi: 10.3390/ani10071211.

Mitchell, S. O. and Rodger, H. D. (2011) A review of infectious gill disease in marine salmonid fish. Journal of Fish Diseases, 34(6), pp. 411–432. doi: 10.1111/j.1365-

2761.2011.01251.x.

Murray, A. G. et al. (2021) A preliminary assessment of indirect impacts on aquaculture species health and welfare in Scotland during COVID-19 lockdown. Veterinary and Animal Science, 11, pp. 1–5. doi: 10.1016/j.vas.2021.100167.

Noble, C., Gismervik, K., Iversen, M. H. et al. (eds.) (2018) Welfare Indicators for farmed Atlantic Salmon: tools for assessing fish welfare http://hdl.handle.net/11250/2575780.

O'Donncha, F. et al. (2021) Data driven insight into fish behaviour and their use for precision aquaculture. Frontiers in Animal Science, 2(July), pp. 1–18. doi: 10.3389/fanim.2021.695054.

Oppedal, F., Dempster, T. and Stien, L. H. (2011) Environmental drivers of Atlantic salmon behaviour in sea-cages: A review. Aquaculture, 311(1–4), pp. 1–18. doi: 10.1016/j.aquaculture.2010.11.020.

Overton, K. et al. (2018) The use and effects of hydrogen peroxide on salmon lice and post-smolt Atlantic salmon. Aquaculture, 486, pp. 246–252. doi: 10.1016/j.aquaculture.2017.12.041.

Overton, K. et al. (2019) Salmon lice treatments and salmon mortality in Norwegian aquaculture: a review. Reviews in Aquaculture, 11(4), pp. 1398–1417. doi: 10.1111/raq.12299.

Paul, E. S. et al. (2020) Towards a comparative science of emotion: Affect and consciousness in humans and animals. Neuroscience and Biobehavioral Reviews, 108, pp. 749–770. doi: 10.1016/j.neubiorev.2019.11.014.

Pettersen, J. M. et al. (2014) Salmon welfare index model 2.0: An extended model for overall welfare assessment of caged Atlantic salmon, based on a review of selected welfare indicators and intended for fish health professionals. Reviews in Aquaculture, 6(3), pp. 162–179. doi: 10.1111/raq.12039.

Pinkiewicz, T. H., Purser, G. J. and Williams, R. N. (2011) A computer vision system to analyse the swimming behaviour of farmed fish in commercial aquaculture facilities: A case study using cage-held atlantic salmon. Aquacultural Engineering, 45(1), pp. 20–27. doi: 10.1016/j.aquaeng.2011.05.002.

Poli, B. M. et al. (2005) Fish welfare and quality as affected by pre-slaughter and slaughter management. Aquaculture International, 13(1–2), pp. 29–49. doi: 10.1007/s10499-004-

9035-1.

Powell, M. D., Reynolds, P. and Kristensen, T. (2015) Freshwater treatment of amoebic gill disease and sea-lice in seawater salmon production: Considerations of water chemistry and fish welfare in Norway. Aquaculture, 448, pp. 18–28. doi: 10.1016/j.aquaculture.2015.05.027.

QSR International Pty Ltd. (2020) Nvivo qualitative data analysis software (March 2020 ed.).

Rey Planellas, S., Little, D. and Ellis, M. (2019) Farmed fish welfare practices : salmon farming as a case study, GAA publications.

Rodger, H. D., Henry, L. and Mitchell, S. O. (2011) Non-infectious gill disorders of marine salmonid fish. Reviews in Fish Biology and Fisheries, 21(3), pp. 423–440. doi: 10.1007/s11160-010-9182-6.

RSPCA (2018) RSPCA welfare standards for farmed Atlantic salmon. Available at: https://science.rspca.org.uk/sciencegroup/farmanimals/standards/salmon (Accessed: 30 October 2022).

RSPCA (2021) RSPCA welfare standards for Farmed Atlantic Salmon, RSPCA welfare standards for Farmed Atlantic Salmon, pp. 1–80. Available at: http://industry.freedomfood.co.uk/media/96636/RSPCA-welfare-standards-for-farmed-Atlantic-salmon-web-version.pdf (Accessed: 30 October 2022).

Saberioon, M. et al. (2017) Application of machine vision systems in aquaculture with emphasis on fish: state-of-the-art and key issues. Reviews in Aquaculture, 9(4), pp. 369–387. doi: 10.1111/raq.12143.

Salmon Scotland (2020) What is RSPCA Assured? Available at: https://www.salmonscotland.co.uk/facts/fish-health-welfare/what-is-rspca-assured (Accessed: 30 October 2022).

Schreck, C. B. and Tort, L. (2016) The concept of stress in fish. Fish Physiology. Academic Press, pp. 1–34. doi: https://doi.org/10.1016/B978-0-12-802728-8.00001-1.

Scottish Sea Farms (2021) Salmon lifecycle - From egg to harvest. Available at: https://www.scottishseafarms.com/sustainability/salmon-lifecycle/ (Accessed: 14 September 2021).

Singer, E., Frankel, M. R. and Glassman, M. B. (1983) The effect of interviewer characteristics and expectations on response. Public Opinion Quarterly, 47(1), pp. 68–83. doi: 10.1086/268767.

Sneddon, L. U., Braithwaite, V. A. and Gentle, M. J. (2003) Novel object test: Examining nociception and fear in the rainbow trout. The Journal of Pain, 4(8), pp. 431–440. doi: 10.1067/S1526-5900(03)00717-X.

SSPO (2021) Monthly Mortality Rate: June 2021, SSPO Lice & Survival Reports. doi: 10.1016/j.ajodo.2021.04.004.

Steinum, T. et al. (2010) Microbial and pathological findings in farmed Atlantic salmon Salmo salar with proliferative gill inflammation. Diseases of Aquatic Organisms, 91(3), pp. 201–211. doi: 10.3354/dao02266.

Stien, L. H. et al. (2013) Salmon Welfare Index Model (SWIM 1.0): A semantic model for overall welfare assessment of caged Atlantic salmon: Review of the selected welfare indicators and model presentation. Reviews in Aquaculture, 5(1), pp. 33–57. doi: 10.1111/j.1753-5131.2012.01083.x.

Størkersen, K. V. et al. (2021) Fish protection during fish production. Organizational conditions for fish welfare. Marine Policy, 129, pp. 1–11. doi: 10.1016/j.marpol.2021.104530.

Superior Fresh (2019) What’s the story behind your supermarket salmon? Available at: https://www.superiorfresh.com/blog-reference/whats-the-story-behind-your-supermarket-salmon (Accessed: 14 September 2021).

Thorstad, E. B. et al. (2015) Effects of salmon lice Lepeophtheirus salmonis on wild sea trout salmo trutta - A literature review. Aquaculture Environment Interactions, 7(2), pp. 91–113. doi: 10.3354/aei00142.

Turnbull, J. F. and Kadri, S. (2007) Safeguarding the many guises of farmed fish welfare. Diseases of Aquatic Organisms, 75(2), pp. 173–182. doi: 10.3354/dao075173.

UN General Assembly (2015) Transforming our world: the 2030 Agenda for Sustainable Development, A/RES/70/1. doi: 10.1163/15718093-12341375.

Weary, D. M. and Fraser, D. (1995) Calling by domestic piglets: reliable signals of need?. Animal Behaviour, 50(4), pp. 1047–1055. doi: 10.1016/0003-3472(95)80105-7.

Webster, J. (2016) Animal welfare: Freedoms, dominions and “A life worth living”. Animals, 6(6), pp. 2–7. doi: 10.3390/ani6060035.

Zhao, J. et al. (2018) Modified motion influence map and recurrent neural network-based monitoring of the local unusual behaviors for fish school in intensive aquaculture. Aquaculture, 493, pp. 165–175. doi: 10.1016/j.aquaculture.2018.04.064.

CHAPTER 3. Farmed salmon welfare practices – insights gained from evaluating standards and farm compliance

Timothy Robert Wiese[1], Sonia Rey Planellas[1], Susan Jarvis[2], Marie Haskell[3], and Jimmy Turnbull[1]

[1]: Institute of Aquaculture, University of Stirling, Stirling, FK9 4LA, UK.
[2]: Global Academy of Agriculture and Food Systems, University of Edinburgh, Midlothian, EH25 9RG, UK.
[3]: SRUC (Scotland's Rural College), West Mains Road, Edinburgh, EH9 3JG, UK.

3.1 Abstract

As concerns for farmed salmon welfare continue to grow, there is an increasing demand for welfare standards to provide additional assurances that farms are meeting their ethical obligations. Despite their importance, few studies have explored the criteria within these welfare standards, and how they are currently implemented within the Scottish salmon farming industry. Furthermore, no study has yet to examine the levels of compliance that farm sites have to these standards, and what insights into their welfare practices this may provide. This study aims to address this knowledge gap by exploring the implementation of a widely adopted set of standards, and assessing Scottish salmon farm sites' compliance to these standards. Pertinent to this study, annual assessments (i.e., audits) were carried out by the accreditors on every certified farm site in order to consistently evaluate their level of compliance. During these audits, a record was made for every instance where a site failed to meet certain requirements (i.e., a 'non-compliance'). From 2011-2019, a total of 1,446 audits were conducted (resulting in an estimated total of 209,000 clauses assessed), from which a total of 1,235 non-compliances were recorded against certified farm sites. Improper record keeping, poor staff training, and incorrect implementation of Veterinary Health and Welfare Plans accounted for more than half of these non-compliances. In general, welfare practices achieved a relatively consistent level of compliance throughout this 9-year period. While containment of fish escapees steadily improved over the years, a growing issue for compliance has been ensuring that proper care is taken during human-animal interactions with the salmon. With regards to the standards themselves, more than 98% of the requirements involved were risk-based, preventative measures for protecting salmon welfare. As a result, only insights into the farms' welfare practices could be obtained from the compliance data. Future iterations of welfare standards should attempt to include direct assessments of physical and behavioural attributes of the salmon so that more direct evidence can be obtained relating to what transpired for their welfare. This study represents the first investigation into the implementation of welfare standards for Scottish farmed salmon and the associated compliance data. These results demonstrate the potential

benefits of collecting and analysing compliance data, as well as the value of such standards adopting a more holistic approach towards monitoring farmed salmon welfare.

Keywords:
Welfare practices; certification; audits; risk analysis; aquaculture

3.2 Introduction

As the salmon farming industry continues to grow, so do concerns amongst stakeholders and the public for maintaining the welfare of farmed salmon (Bergqvist and Gunnarsson, 2013; Barreto *et al.*, 2021; Wiese *et al.*, 2023). There is now ample evidence suggesting that fish are sentient, and therefore capable of perceiving their own welfare (Sneddon, Braithwaite et al., 2003a; Chervova and Lapshin, 2011; Kristiansen *et al.*, 2020). The notion of sentience alone brings forward a number of ethical implications for farmed fish welfare (Ashley, 2007; Huntingford and Kadri, 2014; Barreto *et al.*, 2021).

In Scotland alone, latest estimates show total production of Atlantic salmon reaching a record high of 205,393 tonnes in 2021, with more than 50 million smolts transferred to sea in the same year (Munro, 2022). Improved production efficiency and product quality within aquaculture have also been directly associated with what standards of welfare can be achieved (Southgate and Wall, 2001; FSBI, 2002). The numerous incentives to improve farmed salmon welfare, along with regulation bodies wishing to avoid additional legislative control, have encouraged the Scottish salmon farming sector to adopt various codes of practice, or 'welfare standards' (see Table 3-1 for definitions) that promote salmon welfare beyond what is required by current legislation (FAWC, 2014). Several salmon farming standards now exist, each with their own set of requisite criteria for acquiring certification. Criteria for these standards varies, each with a different emphasis on sustainability, product quality, and fish welfare assurances. The extent to how much farmed salmon welfare is currently valued within the industry is reflected by the number of standards that now include welfare within their criteria (Scottish Salmon Producers Organisation, 2015; Best Aquaculture Practices, 2016; GlobalG.A.P., 2017; RSPCA, 2021; Aquaculture Stewardship Council, 2023).

Certifying around 70% of Scottish salmon farms, the 'RSPCA welfare standards for farmed Atlantic salmon' are specifically aimed towards salmon welfare assurance (Rey Planellas, Little and Ellis, 2019; Salmon Scotland, 2020a; RSPCA, 2021). Their purpose is to act as an auditable set of standards "set at the limit of what is achievable, in terms of animal husbandry and commercial viability" (RSPCA, 2009). That is, they are designed with the aim of protecting against all of the various factors that may negatively impact salmon welfare.

The standards achieve this with a list of numbered requirements (i.e., ‘clauses’) which certified sites must comply with. These clauses involve species-specific requirements that are organised into different sections of the standards by their relevance to certain production stages or husbandry practices (e.g., sections for ‘Management’, ‘Freshwater (pre-smolt/juvenile fish)’, ‘Seawater’, ‘Transport’, etc.) (RSPCA, 2021). The principles of these clauses, which have covered farmed Atlantic salmon since 2006, are originally based upon the ‘Five Freedoms’ as defined by the FAWC (FAO, 2007; RSPCA, 2018a). In addition, the standards outline that these five freedoms will be better maintained if those responsible on-farm practice and provide “caring and responsible management”, “conscientious stockmanship”, “appropriate environmental design”, and “considerate / humane handling, transport, and slaughter” (RSPCA, 2021, pg. 4).

Table 3-1: Working definitions for terms used in this study.

Welfare standards	A list of clauses (i.e., requirements) that farm sites holding the assurance label must comply with. They are designed to ensure the well-being and humane treatment of animals.
Audit	An annual on-farm assessment carried out by the accreditation scheme to assess a farm site’s compliance to the relevant clauses.
Clause = welfare measure / indicator	Any clause is designed to be auditable, and is thus effectively a form of welfare assessment. Since welfare cannot be quantitatively “measured” in the same manner as physical parameters of an animal, the term “measure” is simply used in this study as a more commonly recognised reference to welfare indicators.
Non-compliance (NC)	A farm site’s failure to meet the requirements of a specific clause, which is then recorded during an audit.

By basing the standards on the Five Freedoms, and primarily focusing on the prevention of issues arising in the first place, the majority of clauses in the RSPCA welfare standards mostly take a ‘risk-based’ approach. Put simply, farmed salmon welfare is protected through preventative measures, ensuring that the factors which ultimately impact salmon welfare (e.g., management, husbandry practices, staff training, conditions of equipment and surrounding enclosures, feeding etc.) are properly managed and maintained. This is in contrast to taking an ‘animal-based’ approach, where farmed salmon welfare is instead monitored more directly through physical and/or behavioural indicators of the animals themselves (Hemsworth et al., 2015).

Regardless of the type of clause involved, the responsibility of compliance to the standards (and thereby upholding the assurances set by the standards) inevitably falls upon the accredited farm sites producing the salmon. Annual audits of these farm sites are the main method for ensuring this compliance (RSPCA, 2021). Each audit involves a site visit, carried out by RSPCA Assured assessors, during which the site is assessed against an ‘assessment checklist’ which includes the list of clauses that are specific to the type of site involved (e.g., seawater / freshwater site, on transportation / wellboats, or at harvest stations) (RSPCA Assured, 2023a). This is because certain clauses are only applicable to a certain life stage or context (e.g., during harvests or treatment for alevins etc.). During these annual audits, a record is then made whenever a farm site has violated the requirements for a certain clause (i.e., a "non-compliance") (RSPCA Assured, 2023b).

The growing demand for improvements in salmon welfare, beyond what is simply required by legislation, consequently leads to a growing demand for evidence of what level of standards are being achieved on Scottish farms (Fidra, 2020; Fidra and Best Fishes, 2022). Helping to improve what evidence can be obtained first requires an understanding of what roles the welfare standards and farm site compliance could play in meeting this demand. How well the welfare standards cover the various approaches to monitoring and safeguarding salmon welfare is one aspect of this evidence. Investigating trends and degrees of non-compliances (NCs) over the years may also offer insights, where the levels of compliance can serve as a benchmark for gauging an industry's progress or decline in complying with the standards set in place (EFCA, 2019). Increasing rates of specific NCs over the years may highlight areas that represent increased risks to salmon welfare. Such information could help provide additional guidance for future on-farm management.

The purpose of this study is to therefore investigate whether the RSPCA welfare standards, and associated farm site compliance data, can provide insights into how welfare practices in the Scottish salmon farming industry have changed over the years. This study therefore aims to answer the following research questions:

1) For what areas of the standards is non-compliance the most common? How has the rate of overall non-compliance changed over the years?

1b) Are there any patterns of non-compliance that could potentially indicate recurring or growing issues in welfare practices for farmed salmon?

2) To what extent do the RSPCA welfare standards incorporate clauses that are either risk-based or animal-based? Why might this be the case?

3.3 Materials and methods

3.3.1 Ethical approval

This study was approved through the University of Stirling ethical review process (GUEP: "Legacy EC2020_21 3") prior to commencement of research.

3.3.2 Organisation of clauses and non-compliances

As newer versions of the standards were released every three years, various clauses were either added, removed, moved to different sections, or assigned different clause 'numbers'. Certain clauses were also repeated within different sections of the standards, as they applied to multiple types of sites. This meant that, throughout 2011-2019, the frequencies of NCs against a specific type of clause could not be matched against any single clause number. For example, throughout 2011-2019, the clause requiring that "stocking densities must not exceed standards" had been assigned the following clause numbers; E5.1, FW1.5, W4.4, HP5.12, and SW1.1. In order to reconcile annual summaries for the frequencies of each type of NC over this 9-year period, NCs had to be listed solely against the exact requirements detailed in their corresponding clause (i.e., the number of non-compliances against "stocking densities must not exceed standards" for each year). This allowed for a list of the total number of NCs, raised annually against each corresponding clause, to be combined into a single excel sheet. Clauses that did not exist throughout the entire 9-year period, and could therefore not have any NCs against them during certain years, were assigned an "N/A" under those years where they did not exist. The final spreadsheet, consolidating all of the data mentioned above, had the following columns dedicated to each clause:

- A description of the requirements (i.e., clause) that were failed to be met.
- The type of welfare assessment involved when this specific clause is audited by an RSPCA Assured assessor (e.g., is the clause risk vs. animal-based).
- The number of times a non-compliance was raised against this clause within each year, from 2011-2019 (N/A if the clause was not yet introduced).

Within this spreadsheet, different types of NCs were then further categorised according to the nature of the violation involved (e.g., farm management, husbandry practices, farm conditions, salmon care, or fish escapees) to allow for comparisons across different areas of the standards. A Sankey diagram was created (using SankeyMATIC) to illustrate and further breakdown these categories (Figure 3-1). As this study focused solely on the standards directly relevant to farmed salmon welfare, clauses relating to the welfare of potential predators or cleanerfish, wider environmental impacts, and human health and safety have been excluded. All non-compliance data were anonymised by RSPCA Assured before being provided to us. This was achieved through providing annual summaries of all NCs, categorised by the corresponding checklists from where they were raised (e.g., during seawater vs. freshwater vs. transportation audits with farm IDs removed), from 2011-2019.

3.3.3 Data analysis

For assessing trends in NCs, the initial step was to account for the number of audits conducted by RSPCA Assured Assessors from which these NCs were identified. From each year, examples of the assessment checklists used for each of the different types of audits were obtained. The total number of clauses investigated within each of these different checklists were counted. The total number of clauses were then multiplied by the total number of times those same checklists were used in audits for each year. The cumulative total, across all different checklists, would then provide an estimate of the total number of clauses that were assessed during each year. The total number of clauses assessed during each year were then compared against each other to determine the 'relative investigative effort'. For example, the year with the largest number of clauses would equal 1, whereas another year that had half the number of clauses assessed would equal 0.5. The total number of NCs raised during each year (or within a category of offense, e.g., 'Farm management') were then divided by their corresponding 'relative investigative effort'. This would result in a standardised 'NC rate', thus taking into account the number of audits that took place (Table 3-4). Clauses that were not directly related to salmon welfare were not included in the above calculations.

For each year, a Pearson's correlation test was carried out between the total number of NCs and the total number of site audits, using IBM SPSS Statistics 28 for Windows 10. Trends in the total number of NCs, from 2011-2019, were visually assessed with a linear regression in both the form of raw data and NC rates (standardised by 'relative investigative effort'). Trends in NC rates within the different categories of offenses were also assessed graphically throughout this 9-year period. The NC rates for each different category were expressed by what percentage of the total NC rate they represented within each year.

3.3.4 Classification of clauses

Owing to the numerous clauses that were added / removed from the standards with each new iteration, only the most up-to-date version of the welfare standards was considered relevant for classifying the clauses by whether their requirements (and how they are audited) were risk-based or animal-based (RSPCA, 2021). Irrelevant clauses, such as those regarding cleanerfish or environmental impacts, were again excluded. The specific requirements for each of the 512 relevant clauses were classified, using the criteria in Table 3-2.

Table 3-2: Criteria used for classifying clauses as being either 'risk-based' or 'animal-based'.

Risk-based:	**Animal-based:**
Primary purpose of the clause is to measure / assess the availability of	Primary purpose of the clause is to measure / assess physical or behavioural

resources provided to the salmon, or to minimise certain risks posed.	attributes of the salmon themselves, or whether the salmon show any signs of injury / disease.
The clause refers to environmental parameters, husbandry practices, management / training of farm staff, or conditions of equipment / enclosures that may indirectly impact salmon welfare.	Rather than being a preventative measure, the clause refers to assessing an outcome of treatment (or lack of treatment) towards the salmon. I.e., it is an indicator of prior welfare problems.
A non-compliance to the clause indicates that an increased risk has been posed to salmon welfare.	A non-compliance to the clause indicates that a direct impact to salmon welfare has been identified by the auditor.

3.4 Results

3.4.1 Audits and number of clauses assessed

The total number of audits carried out varied from one year to another (Table 3-3). Of the 1,446 total audits, 59% (n=857) were carried out on seawater sites, 30% (n=436) on freshwater sites, 9% (n=125) on transportation / wellboats, and 2% (n=28) on harvest stations.

Table 3-3: Total number of site assessments carried out annually, from 2011-2019, on all RSPCAA certified Scottish salmon farm sites (including seawater sites, freshwater / hatchery sites, transportation / wellboats, and harvest stations). Numbers with a * were interpolated based on relative proportions from other years, due to only general audit numbers being provided for these years.

Year:	Seawater audits	Freshwater audits	Transportation / wellboat audits	Harvest station audits	Total number of site audits
2011	127	86	13	6	232
2012	123	55	13	7	198
2013	110	54	12	4	180
2014	94*	48*	14*	4*	160
2015	79*	40*	11*	4*	134
2016	116	48	12	5	176
2017	49	26	5	3	83
2018	53	35	8	2	98
2019	106	44	31	4	185
Total	857	436	119	39	1446

3.4.2 Non-compliances by nature of offense

Throughout 2011-2019, there were a total of 1,235 NCs, with a mean 137 NCs per year. Figure 3-1 provides a breakdown of these NCs, categorised by their nature of offense (and the number of NCs associated within each category / sub-category). The majority of NCs related to 'Farm management' (n = 651, ~53% of all NCs). Clauses within this category covered requirements for the correct implementation of the Veterinary Health and Welfare Plan (VHWP), staff training on fish welfare, and maintaining sufficient records of husbandry practices, equipment maintenance, and conditions on-site. The clause with the second highest number of NCs was also under the 'Farm management', with 91 NCs for 'Staff not familiar with RSPCA standards / standards not part of site induction' (Table 3-5). Within this largest category of total NCs, 38% (n=253) of the NCs were related to improper record keeping.

Table 3-4: Standardisation of non-compliance (NC) rates by relative investigative effort, calculated by total number of clauses

	2011	2012	2013	2014	2015	2016	2017	2018	2019	Total
Total no. of NCs	233	241	119	66	63	93	135	128	157	1235
Total no. of clauses assessed:	33,632	28,677	25,986	23,348	19,823	24,472	11,533	14,364	27,209	209,044
Relative investigative effort:	1	0.85	0.77	0.69	0.59	0.73	0.34	0.43	0.81	N/A
"NC rates" (standardised):	233	283	154	95	107	128	394	300	194	N/A

The clause with the highest number of NCs was in 'Farm conditions' (n=182, ~15% of all NCs), with 99 NCs for 'stocking densities exceeds the standards' (Table 3-5). NCs relating to 'Husbandry practices' accounted for ~21% of all NCs (n=253), the majority of which consisted of dealing with mortalities (n=106), and crowding / grading / transport practices (n=71). NCs relating to 'Salmon care' accounted for ~8% of all NCs (n=101), 28% of which

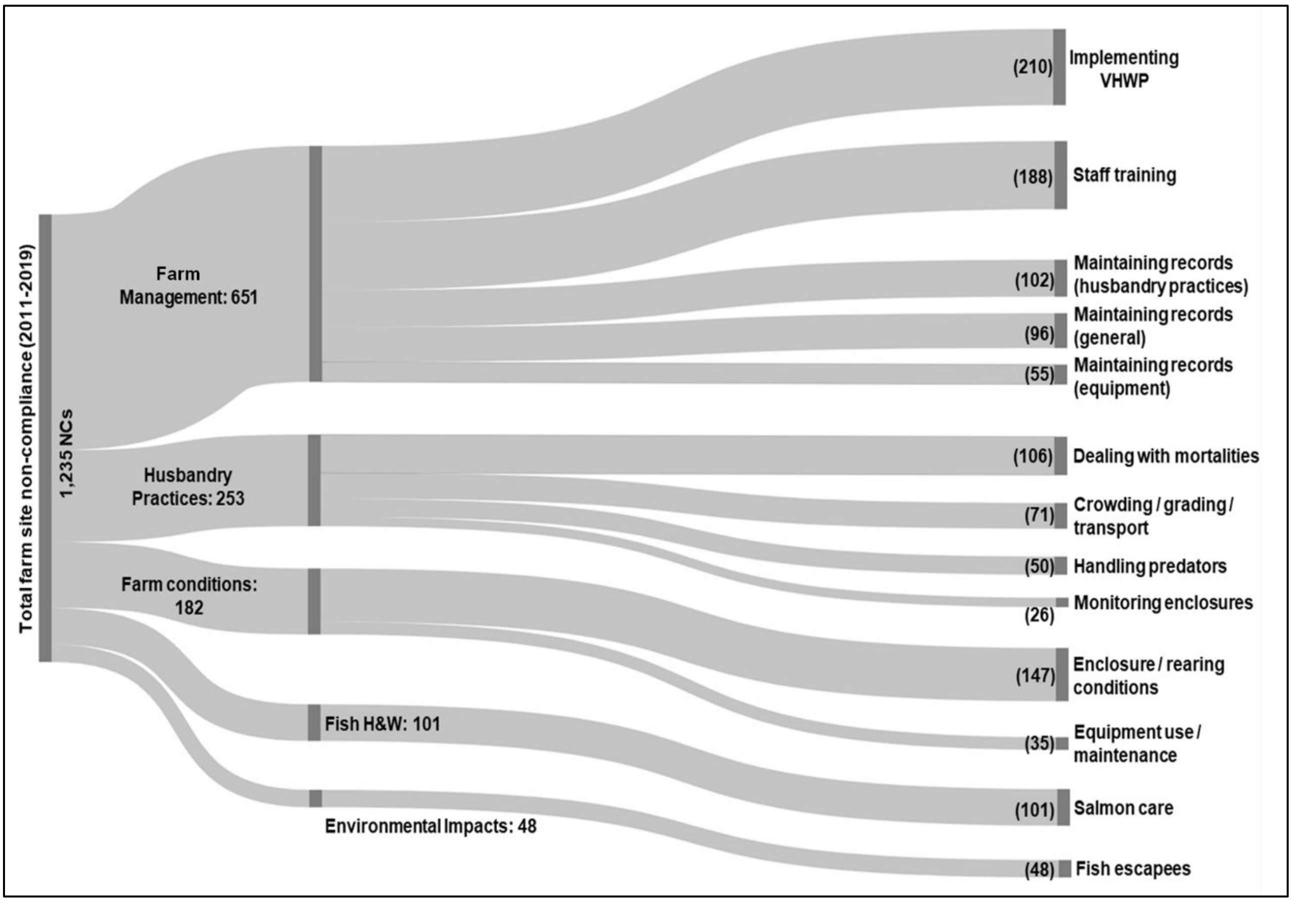

Figure 3-1. Sankey diagram (created using SankeyMATIC) illustrating a breakdown of all non-compliances raised (1235) throughout 2011-2019 during RSPCAA site audits, for which posed an increased risk to farmed salmon. Non-compliances (NCs) are categorised by nature of offense (farm management, husbandry practices, farm conditions, salmon care, fish escapees).

related to the more severe, animal-based clauses for the salmon (e.g., recurring physical damage to the fish observed, seriously injured / diseased fish not humanely killed without delay, fish showing obvious signs of stress prior to harvest etc.).

Table 3-5. Top 10 clauses by number of non-compliances (NCs) raised against them throughout 2011-2019 during RSCPAA site assessments, alongside their corresponding nature of offense.

Category (by nature of offense)	Clause within the RSPCA welfare standards	Total NCs (2011-2019):
Farm conditions (poor rearing conditions)	Stocking density exceeds the standards	99
Management (staff training)	Staff not familiar with standards content	91
Management (records)	Full inspection records not maintained or in place on site	59
Management (staff training)	Manager has not attended a fish welfare course, or staff not adequately trained for fish husbandry and welfare	56
Management (VHWP)	VHWP does not show remedial action taken to mitigate welfare issues on site	54
Husbandry practice (mortalities)	Removal of dead fish does not take place min. twice weekly	53
Management (VHWP)	No site-specific VHWP in place, or VHWP not being regularly updated / implemented	48
Husbandry practice (crowding)	Oxygen levels not monitored and recorded throughout all crowding operations	48
Husbandry practice (mortalities)	Cause of fish death not classified	41
Management (VHWP)	Grading plan not part of VHWP / not on site	35

3.4.3 Trends in non-compliance over the years

Regression analysis on the total number of NCs each year suggested at first a slight negative correlation with time and that, overall, NCs were decreasing over the years (Figure 3-2). However, when the total number of audits carried out each year were taken into account, the annual trend in 'NC rates' showed that there was no consistent pattern in the overall rate of non-compliance throughout the 2011-2019 period (Figure 3-2). No significant association was initially found between total number of site audits and overall NCs raised each year throughout 2011-2019 (Pearson correlation = 0.523, $p = 0.148$). However, between 2017-2018, the average number of site audits were significantly fewer (approximately 50%) in comparison to other years, while having on average more than double the NC rates. If 2017 and 2018 are treated as outliers, a significant association was found between number of site audits and overall NCs raised each year (Pearson correlation = 0.878, $p < 0.01$). While there was no consistent pattern in overall NC rates throughout 2011-2019, trends in certain categories were more apparent.

On average, NC rates relating to matters of 'Salmon care' accounted for ~5% of total NC rates from 2011-2013, ~9% from 2014-2016, and ~11% from 2017-2019 (Figure 3-3). Conversely, NC rates relating to fish escapees have, on average, accounted for ~4% of total

NC rates from 2011-2013, and ~2% from 2014 onwards (Figure 3-3). All remaining categories have shown no consistent patterns throughout the years.

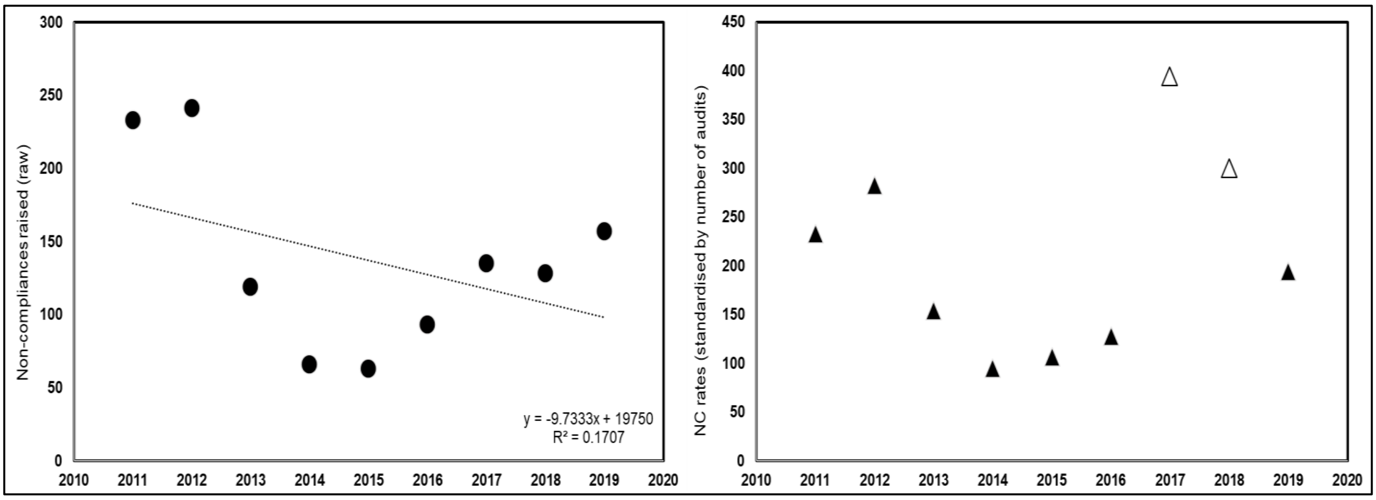

Figure 3-2. Annual non-compliance (NC) data from 2011-2019. Black circles indicate the raw data for total NCs raised. Black triangles indicate NC rates, standardised by number of audits carried out for each year. White triangles highlight 2017 and 2018 as potential outliers. A trendline of raw NC data is also shown to illustrate a potential trend that, without the removal of 2017 and 2018 data, no longer exists in the standardised NC rates.

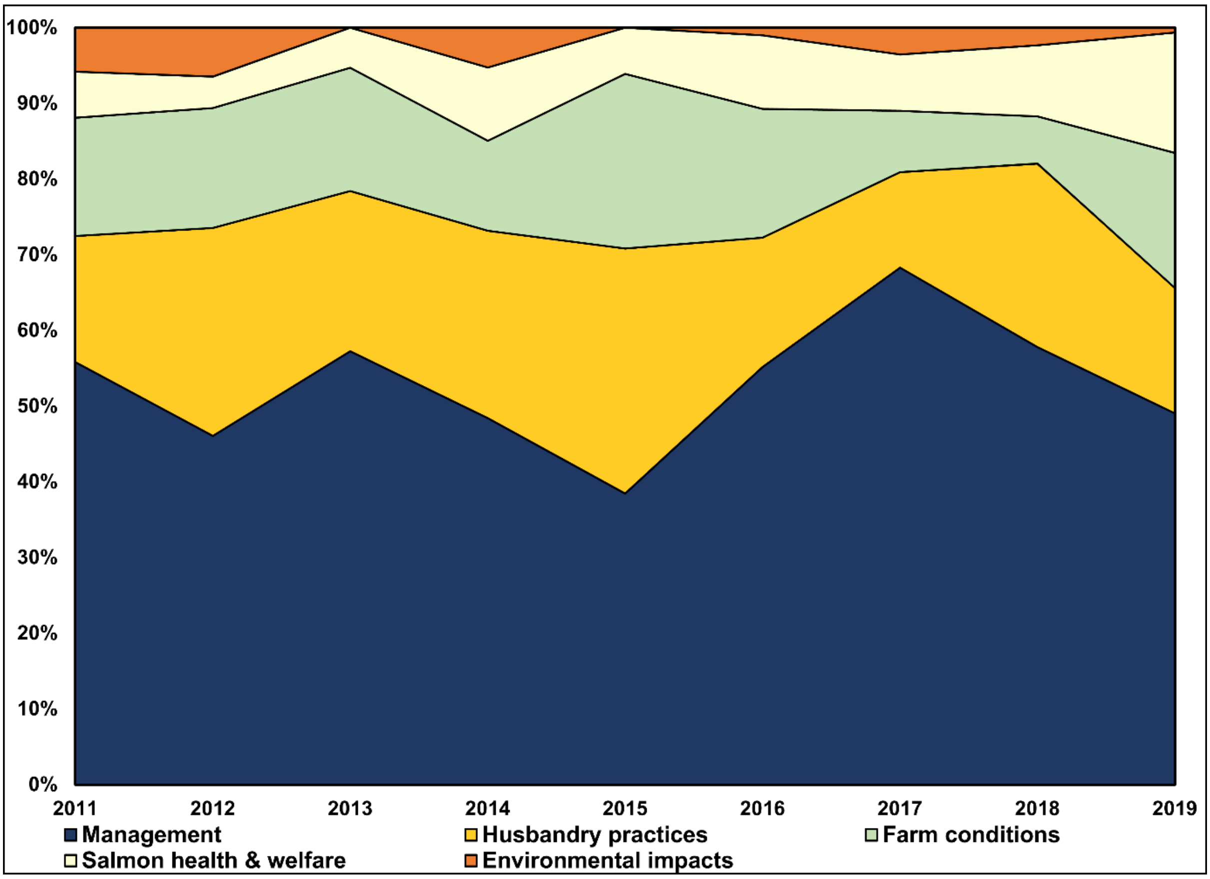

Figure 3-3. Trends in non-compliance (NC) rates on an annual basis, across 5 different categories of clauses that share a common type of offense. NC rates for each category are represented as the percentage proportion of total NCs raised within each year.

3.4.4 How many clauses are risk-based vs. animal-based

Approximately 2% of all clauses within the 2021 welfare standards could arguably function as animal-based measures when being audited (10 out of 512 clauses examined, Table 3-6).

Table 3-6. The 10 clauses from the RSPCA Assured welfare standards (2021) that arguably function as animal-based measures during audits.

Clause	Requirement(s)
H 1.4	There must be no recurring physical damage occurring on fish attributable to features of their environment, husbandry procedures, or unrecognised disease challenge.
H 1.7, FW 9.29, H 4.7	Any fish suffering from overt physical damage, or disease symptoms (incl. sea lice), must be segregated & treated humanely without delay.
H 2.3, HP 1.6, FW 9.25	Under no circumstances must seriously injured or sick fish be left to die in the air.
H 5.1	Mutilations involving the removal of sensitive tissue are prohibited.
H 5.2	Marking methods that cause distress or injury to fish must not be used.
HP 8.2	Fish must not have been produced by breeding techniques that result in health or welfare problems.

3.5 Discussion

Outside of legislation, the sole avenue for providing assurances on farmed salmon welfare are through what standards are set in place (FAWC, 2014), and the extent to which accredited farm sites comply with these standards. Farm sites are incentivised to be certified under these welfare standards because of consumer demand, with past Eurobarometer surveys indicating that up to 72% of the British public are willing to pay more for higher welfare standards (European Commission, 2016). The quality of these welfare assurances are directly linked to how well the standards cover the various aspects of monitoring, and ultimately safeguarding, salmon welfare. Trends and degrees of non-compliance from the farms can then also provide insights into what welfare practices may be improving or deteriorating, and where the most common issues arise with compliance.

The results from this study represent nine years of non-compliance data from Scottish salmon farms certified under the RSPCA Assured welfare standards. Overall NC rates suggest that general welfare practices for accredited farm sites have remained relatively constant throughout 2011-2019. Throughout this period, improper management contributed the most towards overall non-compliance, accounting for more than half of all NCs. NCs relating to poor record keeping, improper staff training on salmon welfare, and incorrect implementation of the Veterinary Health and Welfare Plan (VHWP) represent the most common factors posing an increased risk to salmon welfare through 2011-2019. Staff training and knowledge of salmon welfare have been ranked as some of the most important factors for farmed salmon welfare (Brown *et al.*, 2008; Berrill *et al.*, 2012). Conversely, competent farm personnel carrying out daily tasks have also been considered to make the most substantial contribution towards protecting fish welfare, through routine interactions / tasks of feeding, cleaning, monitoring, and treating the fish (Størkersen *et al.*, 2021). With regards to husbandry practices and general farm conditions in the current study, the improper handling of mortalities and inappropriate stocking densities were the most frequent NCs respectively. Severe violations to the standards, where obvious mistreatment of the salmon and consequent suffering was observed, were far less frequent.

When investigating what these NCs may indicate about how overall farm welfare practices have changed over the years, additional factors must be considered. This includes the comparative likelihood of certain clauses being violated over others; if NCs are more likely to occur for one clause than another, then simply comparing the frequency of NCs between two clauses (or category of clauses) becomes less meaningful.

In instances of less severe NCs (i.e., those not likely to cause suffering to the salmon), the RSPCA Assured provide a formal warning to the farm staff involved, who are then obligated to resolve any identified NCs or risk having their certification suspended (RSPCA Assured, 2023b). However, if more severe NCs are identified, (i.e., RSPCA Assured notes that salmon have likely suffered directly as a result of neglect / malpractice), further investigations may take place that could lead to immediate withdrawal of the RSPCA Assured label (RSPCA Assured, 2023b). There is therefore more incentive for farms to avoid the most severe non-compliances. These more “serious”, animal-based clauses found within the standards also form less than 2% of all clauses, thereby limiting the total number of NCs that could possibly be identified during audits. Severe violations to the standards are thus expected to form a minority of the total NCs. In contrast, there are significantly more clauses involved in record keeping, with the consequences of violating them likely not being as severe for the farms. Differences in the overall non-compliance trends over the years (when comparing “raw” number of NCs vs standardised NC rates) indicate that the number of NCs raised are at least partially linked to the number of audits that took place in that same year. Assessing trends in NCs, either between different years or across different sections of the standards, cannot be done reliably without accounting for the ‘relative investigative effort’ involved when these NCs were identified (Sutinen, Rieser and Gauvin, 1990; Bergseth, Russ and Cinner, 2015; Read, West and Kelaher, 2015). In other words, an increased number of NCs could simply be due to the corresponding clause being audited more. This means that direct comparisons of “raw” numbers of NCs between different areas of the standards cannot be meaningfully assessed. Instead, it is more appropriate to examine the trends of NC rates across these different categories, and compare how they have changed over the years.

There have been no consistent patterns in NC rates regarding management, husbandry practices, or farm conditions, which formed the majority of the standards throughout 2011-2019. However, there were more apparent trends in more specific areas of the standards. Welfare practices relating to fish escapees have steadily improved over the years, whereas practices relating to salmon care have steadily deteriorated. This steady increase in NC rates relating to salmon care includes the few animal-based clauses currently present in the standards, such as “any fish suffering from overt physical damage, or disease symptoms, must be segregated and treated humanely without delay”. However, the vast majority of clauses in this category are still risk-based, focusing on what care must be taken by farm staff for ensuring humane handling, interventions, fasting, and culling.

To shed light on the unusual number of audits and NC rates observed between 2017-2018, RSPCA Assured staff provided supplementary information to offer potential explanations. In 2016, a large company withdrew from the RSPCA Assured scheme. This lead to ~60 sites that were no longer audited from 2017 onwards, which could help explain the following years having the lowest ‘relative investigative effort’ from 2011-2019. In addition, a new version of the standards was implemented in early 2018. From 2011 onwards, 35% of newly introduced clauses were first included in the 2018 standards. This may have contributed to higher NC rates until the farm staff became adjusted to these new clauses.

Certain clauses have also not been consistently audited, as they would not always be applicable to certain assessment checklists or even certain sites. For example, all seawater sites in 2011 would likely not have had the exact same clauses audited (e.g., clauses such as M3.12 requiring the monitoring of smolts). Even within the same site over many years, specific clauses may only apply for the years where certain events coincided with the annual audit (e.g., a predator attack). The lack of such standardisation, both within and between certification schemes, is a significant complication that results in a lack of clarity and potentially impedes demand for products (Main *et al.*, 2014). Ideally, audits could record (onto a private, aggregated database) every instance of a clause being assessed. This would allow for reliable calculations of NC rates for every clause. Flexible comparisons of NC rates would then be possible between any groups of clauses, regardless of the ‘relative investigative effort’ involved.

The current non-compliance data allows for an investigation into the welfare practices of Scottish salmon farms, comparing how different risks have been minimised as well as highlighting potential key areas for improving compliance (i.e., management and salmon care). However, without the appropriate inclusion of animal-based clauses for the salmon, this data is not yet able to provide a more complete, outcome-based story of what level of welfare was achieved for the salmon. This is largely due to the majority of clauses adopting a risk-based approach. There are costs and benefits to implementing either risk-based or animal-based clauses within any welfare standards. Either approach requires considerations of practicality (i.e., can these clauses be feasibly assessed during an audit without disturbing the salmon or farm staff's routines?) as well as effectiveness (i.e., how well do these clauses help provide a definitive overview of salmon welfare?). There are many advantages to adopting a risk-based approach. Fish farming in densely populated, confined enclosures increases the risk of outbreaks of highly infectious diseases which can intensify rapidly before they are noticed (Robertsen, 2011; Gjessing *et al.*, 2017; Buchmann, 2022). Without routine invasive sampling of individual salmon, asymptomatic infections can continue spreading while remaining undetected for extended periods of time (Hiney, Kilmartin and Smith, 1994; Morton and Routledge, 2016). The more advanced an outbreak (e.g., of AGD), the more difficult they can be to treat (Rodger, 2014). Even after detection, various treatments often severely impact salmon health and incur significant to the farms (Liu and Bjelland, 2014; Hjeltnes *et al.*, 2017). Animal-based clauses are inherently retrospective, often only detecting salmon that are already diseased or injured (Noble *et al.*, 2018). In contrast, risk-based clauses are implemented to ensure optimal rearing conditions are maintained, thereby minimising issues from arising in the first place. 'Prevention over treatment' is often regarded as the more effective, and less costly, form of health (and welfare) management (Noble *et al.*, 2018; Barrett *et al.*, 2020). Clauses that are animal-based also have the difficult task of ensuring that a representative sample of the fish is obtained, so that the "true" situation within an enclosure is observed (Noble *et al.*, 2018; Wiese *et al.*, 2023). Such measures often involve a time-consuming, laborious process of manually handling the fish, putting added stress on the farm staff's responsibilities and the salmon themselves (Noble *et al.*, 2018). In contrast, risk-based measures are often relatively easy and inexpensive to monitor or audit on-site (Noble *et al.*, 2018); for example, clauses that require appropriate record keeping, staff training, or equipment maintenance can be audited with simple visual inspections or interviews.

However, minimising risks is only one aspect of safeguarding animal welfare. Even when perfect risk-management and "optimal" conditions are perceived to be set in place, there is no guarantee that issues will not arise. The Brambell report, which led to the formation of the Five Freedoms (for which the RSPCA Assured welfare standards are based upon), stresses how welfare assessments must attempt to integrate any available physical and/or behavioural indicators of an animal's welfare (Brambell *et al.*, 1965; Elischer and Conklin, 2019). Such indicators are essential for providing evidence for what levels of welfare were achieved for the salmon, rather than solely providing assurances on what risks were mitigated. There has also been a growing opinion that welfare should be more than simply the absence of suffering (Mench, 1998; Mellor, 2012, 2016; Fife-Cook and Franks, 2019; Barreto *et al.*, 2021). A solely risk-based approach tends to focus largely on the elimination of risks to animal welfare, often leaving out any potential ‘positive’ aspects of welfare as a result (Fife-Cook and Franks, 2019).

The current RSPCA Assured welfare standards do provide additional guidelines for monitoring a range of animal-based measures that “should” be examined during various practices, including a ‘summary of observations during fish slaughter’, ‘scoring systems for deformities and injuries of fins, scales, spine, snout, jaw, eye, and operculum’, and ‘crowd intensity scales’ (RSPCA, 2021). However, these guidelines have not yet been drawn up into clauses for which farms are required to comply with. The newest version of the standards now states that direct measures of animal welfare are essential to understanding “what levels of welfare are being achieved, and therefore better understanding what impact the resources being provided (and management practices being implemented) are having on the animals” (RSPCA, 2021, pg. 49). The RSCPA further recognises the importance of animal-based measures by stating their intention to formally include them as clauses in the next iteration of the standards (RSPCA, 2021, pg. 49).

The RSPCA Assured are not alone in recognising the value of animal-based measures for farmed salmon welfare. During a stakeholder meeting in collaboration with the DEFRA Innovation centre, the integration and application of behavioural and physiological indices was voted as the fourth most important area of development for farmed fish welfare in the UK (Berrill *et al.*, 2012). Considering the demand for implementing more animal-based measures into welfare assessment schemes, the aforementioned limitations commonly found within this class of measures highlights a clear need for further developing animal-based measures that

1) Can provide early warning signs for health and welfare issues that arise, giving farm staff adequate time to carry out the necessary interventions
2) Are non-intrusive and practical in their use, having little to no impact on either salmon welfare or farm staff’s routines
3) Have simple avenues for which automation can be built upon them for further improve their practicality and effectiveness

The successful implementation of such measures would then help improve welfare standards in two manners:

1) Facilitate the inclusion of clauses that attempt to monitor aspects of welfare that cannot be covered through a risk-based approach, namely those which promote positive welfare as a motivational framework within the standards (Webster, 2016). This could include behavioural assessments that allow for inferences to be made on the emotional state of the animals (Mellor, 2012)

2) Help improve the auditability of the standards through inclusion of clauses that may, in the future, be monitored and assessed directly through non-invasive and remote monitoring. Further developments in automation and computer vision that improve monitoring and data collection could someday allow these measures to require minimal additional time or effort (from farm staff or auditors) for ensuring compliance (Rey Planellas, Little and Ellis, 2019; Erp-van der Kooij and Rutter, 2020; Yang *et al.*, 2021).

3.6 Conclusion

From 2011-2019, welfare practices within the Scottish salmon farming sector have achieved relatively consistent levels of compliance to the RSPCA Assured welfare standards. Although welfare practices related to staff training, record keeping, and correct implementation of the VHWP have remained consistent over the years, these are key areas of focus for improving overall compliance. Welfare practices relating to farm conditions and husbandry practices would generally benefit the most by maintaining more appropriate stocking densities and improving how mortalities are dealt with. Containment of fish escapees have been steadily improving. While not representative of farm staff conduct overall, a growing issue for compliance is ensuring proper care is taken to minimise risks posed during interactions with the salmon. Improving such welfare practices may also be related to staff training.

This study also highlighted that the extent of information that can be derived from compliance data largely depends on what clauses the standards consist of. For welfare standards prioritising a risk-based approach, only insights towards the farms' welfare practices can be obtained. With the RSPCA's intention to include more animal-based measures as clauses in the upcoming versions of the standards, compliance data in future years may be able to provide a more complete (and direct) overview as to what transpired for the welfare of the farmed salmon.

There is a clear demand for including animal-based measures in welfare standards which hold farms accountable for ensuring that salmon welfare is definitively being safeguarded to an acceptable level. True accountability within a welfare assessment scheme (or set of standards) requires the integration of both risk-based and animal-based measures, so that the assessments / audits involved can reflect the degree of welfare achieved that is closest to the animals themselves. For farm staff and accreditors alike, there is a need for further development of animal-based measures that are both non-intrusive and practical so that their use for on-farm monitoring or auditing becomes a realistic prospect.

3.7 Acknowledgements

The authors would like to express their gratitude to the RSPCA Assured team and Malcolm Johnstone for generously providing all of the anonymised compliance data from 2011 to 2019. This publication reflects solely the views of the authors.

3.8 References

Aquaculture Stewardship Council (2023) ASC Salmon Standard Version 1.4, Farm Standards. Available at: https://www.asc-aqua.org/what-we-do/our-standards/farm-standards/the-salmon-standard/.

Ashley, P. J. (2007) Fish welfare: Current issues in aquaculture. Applied Animal Behaviour Science, 104(3–4), pp. 199–235. doi: 10.1016/j.applanim.2006.09.001.

Barreto, M. O., Rey Planellas, S., Yang, Y. et al. (2021) Emerging indicators of fish welfare in aquaculture. Reviews in Aquaculture, 14(1), pp. 1–19. doi: 10.1111/raq.12601.

Barrett, L. T., Oppedal, F., Robinson, N. et al. (2020) Prevention not cure: a review of methods to avoid sea lice infestations in salmon aquaculture. Reviews in Aquaculture, 12(4), pp. 2527–2543. doi: 10.1111/raq.12456.

Bergqvist, J. and Gunnarsson, S. (2013) Finfish Aquaculture: Animal Welfare, the Environment, and Ethical Implications. Journal of Agricultural and Environmental Ethics, 26(1), pp. 75–99. doi: 10.1007/s10806-011-9346-y.

Bergseth, B. J., Russ, G. R. and Cinner, J. E. (2015) Measuring and monitoring compliance in no-take marine reserves. Fish and Fisheries, 16(2), pp. 240–258. doi: 10.1111/faf.12051.

Berrill, I. K., Cooper, T., MacIntyre, C.M. et al. (2012) Achieving consensus on current and future priorities for farmed fish welfare: A case study from the UK. Fish Physiology and Biochemistry, 38(1), pp. 219–229. doi: 10.1007/s10695-010-9399-2.

Best Aquaculture Practices (2016) Aquaculture Facility Certification - Salmon Farms, BAP Standards & Guidelines. Available at: https://www.bapcertification.org/Standards.

Brambell, F. W. R. (1965) Report of the Technical Committee to Enquire into the Welfare of Animals kept under Intensive Livestock Husbandry Systems. London: HMSO.

Brown, D. M. et al. (2008) Animal welfare aspects of husbandry systems for farmed Atlantic salmon - Scientific Opinion of the Panel on Animal Health and Welfare. The EFSA Journal, 6(7), pp. 736. doi: 10.2903/j.efsa.2008.796.

Buchmann, K. (2022) Control of parasitic diseases in aquaculture. Parasitology, 149(14), pp. 1985-1997. doi: 10.1017/S0031182022001093.

Chervova, L. S. and Lapshin, D. N. (2011) Behavioral Control of the Efficiency of Pharmacological Anesthesia in Fish. Journal of Ichthyology, 51(11), pp. 1126–1132. doi: 10.1134/S0032945211110026.

EFCA (2019) Compliance Evaluation. Available at: https://www.efca.europa.eu/en/content/compliance-evaluation (Accessed: 30 October 2021).

Elischer, M. and Conklin, T. (2019) The Five Freedoms: A history lesson in animal care and welfare, Michigan State University Extension. Available at: https://www.canr.msu.edu/news/an_animal_welfare_history_lesson_on_the_five_freedoms (Accessed: 8 February 2023).

Erp-van der Kooij, E. V. and Rutter, S. M. (2020) Using precision farming to improve animal welfare, CAB Reviews: Perspectives in Agriculture. Veterinary Science, Nutrition and Natural Resources, 15(51). doi: 10.1079/PAVSNNR202015051.

European Commission (2016) Attitudes of Europeans towards Animal Welfare - Special Eurobarometer. Available at: https://europa.eu/eurobarometer/surveys/detail/2096 (Accessed 10 January 2023).

FAO (2007) Animal welfare and ‘free-range’ schemes. Available at: http://www.fao.org/3/ai388e/AI388E21.htm (Accessed 30 October 2021).

FAWC (2014) Opinion on the Welfare of Farmed Fish. *FAWC report,* pp. 1-40. Available at: https://assets.publishing.service.gov.uk/media/5a7eb32340f0b62305b829c4/Opinion_on_the_welfare_of_farmed_fish_at_the_time_of_killing.pdf (Accessed: 11 October 2021).

Fidra (2020) Fidra survey reveals: UK consumers will pay more for ‘sustainably produced’ Scottish salmon & feel ill-informed, Best Fishes. Available at: https://www.bestfishes.org.uk/fidra-survey-reveals-uk-consumers-will-pay-more-if-scottish-salmon-is-sustainably-produced-but-dont-feel-well-informed/ (Accessed: 3 June 2023).

Fidra and Best Fishes (2022) Scottish Salmon Farming - Consumer survey results. Available at: https://www.bestfishes.org.uk/wp-content/uploads/Consumer-survey-2021-Report_FINAL.pdf (Accessed: 3 June 2023).

Fife-Cook, I. and Franks, B. (2019) Positive welfare for fishes: rationale and areas for future study. Fishes, 4(2), p. 31. doi: 10.3390/fishes4020031.

Smith, V.J. (2002) Fish Welfare, Briefing Paper 2, Fisheries Society of the British Isles (FSBI). Fish & Shellfish Immunology, 3(10), pp. 213-214. doi: 10.1006/fsim.1999.0251.

Gjessing, M. C., Thoen, E., Tengs, T. et al. (2017) Salmon gill poxvirus, a recently characterized infectious agent of multifactorial gill disease in freshwater- and seawater-reared Atlantic salmon. Journal of Fish Diseases, 40(10), pp. 1253–1265. doi: 10.1111/jfd.12608.

GlobalG.A.P. (2017) The GLOBALG.A.P. Integrated Farm Assurance (IFA) Standard. Available at: https://www.globalgap.org/uk_en/documents/#fq=gg.document.type:(%22checklist%22+OR+%22regulations%22+OR+%22cpacc%22)&fq=gg.standard.gg:(%22ifa5%22)&fq=gg.subscope:(%22aquacullture%22)&fq=con_locales:(%22en%22) (Accessed: 29 January 2020).

Hemsworth, P. H., Mellor, D.J., Cronin, G.M. et al. (2015) Scientific assessment of animal welfare. New Zealand Veterinary Journal, 63(1), pp. 24–30. doi: 10.1080/00480169.2014.966167.

Hiney, M. P., Kilmartin, J. J. and Smith, P. R. (1994) Detection of *Aeromonas salmonicida* in Atlantic salmon with asymptomatic furunculosis infections. Diseases of Aquatic Organisms, 19(3), pp. 161–167. doi: 10.3354/dao019161.

Hjeltnes, B., Bang-Jensen, B., Bornø, G. et al. (2017) The health situation in Norwegian aquaculture 2016, Norwegian Veterinary Institute, p. 127.

Huntingford, F. A. and Kadri, S. (2014) Defining, assessing and promoting the welfare of farmed fish. OIE Revue Scientifique et Technique, 33(1), pp. 233–244. doi: 10.20506/rst.33.1.2286.

Kristiansen, T. S. (2020) The welfare of fish, Diseases of Aquatic Organisms, 75(2), pp. 155-159. doi: 10.3354/dao075155.

Liu, Y. and Bjelland, H. V. (2014) Estimating costs of sea lice control strategy in Norway. Preventive Veterinary Medicine, 117(3–4), pp. 469–477. doi: 10.1016/j.prevetmed.2014.08.018.

Main, D. C. J., Mullan, S., Atkinson, C. et al. (2014) Best practice framework for animal welfare certification schemes. Trends in Food Science and Technology, 37(2), pp. 127-136. doi: 10.1016/j.tifs.2014.03.009.

Mellor, D. J. (2012) Animal emotions, behaviour and the promotion of positive welfare states. New Zealand Veterinary Journal, 60(1), pp. 1–8. doi: 10.1080/00480169.2011.619047.

Mellor, D. J. (2016) Updating animal welfare thinking: moving beyond the “Five Freedoms” towards “A Life Worth Living”. Animals, 6(3), pp. 1–20. doi: 10.3390/ani6030021.

Mench, J. (1998) Thirty years after Brambell: whither animal welfare science?. Journal of Applied Animal Welfare Science, 1(2), pp. 91–102. doi: 10.1207/s15327604jaws0102_1.

Morton, A. and Routledge, R. (2016) Risk and precaution: Salmon farming, Marine Policy, 74, pp. 205–212. doi: 10.1016/j.marpol.2016.09.022.

Munro, L. A. (2022) Scottish Fish Farm Production Survey 2021. Available at: https://www.gov.scot/publications/scottish-fish-farm-production-survey-2021/pages/5/ (Accessed: June 30, 2023).

Noble, C., Gismervik, K., Iversen, M. H. et al. (eds.) (2018) Welfare Indicators for farmed Atlantic Salmon: tools for assessing fish welfare http://hdl.handle.net/11250/2575780.

Read, A. D., West, R. J. and Kelaher, B. P. (2015) Using compliance data to improve marine protected area management. Marine Policy, 60, pp. 119–127. doi: 10.1016/j.marpol.2015.06.008.

Rey Planellas, S., Little, D. and Ellis, M. (2019) Farmed fish welfare practices: salmon farming as a case study, GAA publications.

Robertsen, B. (2011) Can we get the upper hand on viral diseases in aquaculture of Atlantic salmon?. Aquaculture Research, 42(SUPPL. 1), pp. 125–131. doi: 10.1111/j.1365-2109.2010.02671.x.

Rodger, H. D. (2014) Amoebic gill disease (AGD) in farmed salmon (*Salmo salar*) in Europe’. Fish Veterinary Journal, 14, pp. 16–27.

RSPCA (2009) RSPCA welfare standards, Farm animal welfare standards. Available at: https://science.rspca.org.uk/sciencegroup/farmanimals/standards (Accessed: 6 December 2022).

RSPCA (2018) RSPCA welfare standards for farmed Atlantic salmon. Available at: https://science.rspca.org.uk/sciencegroup/farmanimals/standards/salmon (Accessed: 28 October 2019).

RSPCA (2021) ‘RSPCA welfare standards for Farmed Atlantic Salmon’, RSPCA welfare standards for Farmed Atlantic Salmon, (February), pp. 1–80. Available at:

http://industry.freedomfood.co.uk/media/96636/RSPCA-welfare-standards-for-farmed-Atlantic-salmon-web-version.pdf (Accessed: 3 June 2023).

RSPCA Assured (2023a) Farm assessments, About RSPCA Assured. Available at: https://www.rspcaassured.org.uk/about-us/farm-assessments/ (Accessed: 3 June 2023).

RSPCA Assured (2023b) Sanctions guidelines, About RSPCA Assured. Available at: https://www.rspcaassured.org.uk/about-us/sanctions-guidelines/ (Accessed: 3 June 2023).

Salmon Scotland (2020) What is RSPCA Assured?. Fish Health & Welfare. Available at: https://www.salmonscotland.co.uk/facts/fish-health-welfare/what-is-rspca-assured (Accessed: 11 January 2023).

Scottish Salmon Producers Organisation (2015) Code of Good Practice for Scottish Finfish Aquaculture. Available at: http://thecodeofgoodpractice.co.uk/chapters/ (Accessed: 15 November 2019).

Sneddon, L. U., Braithwaite, V. A. and Gentle, M. J. (2003) Do fishes have nociceptors? Evidence for the evolution of a vertebrate sensory system. Proceedings of the Royal Society B: Biological Sciences, 270(1520), pp. 1115–1121. doi: 10.1098/rspb.2003.2349.

Southgate, P. and Wall, T. (2001) Welfare of farmed fish at slaughter. In Practice, 23, pp. 277–284.

Størkersen, K. V., Osmundsen, T. C., Stien, L. H. et al. (2021) Fish protection during fish production. Organizational conditions for fish welfare. Marine Policy, 129, pp. 1–11. doi: 10.1016/j.marpol.2021.104530.

Sutinen, J. G., Rieser, A. and Gauvin, J. R. (1990) Measuring and explaining noncompliance in federally managed fisheries. Ocean Development and International Law, 21(3), pp. 335–372. doi: 10.1080/00908329009545942.

Webster, J. (2016) Animal welfare: Freedoms, dominions and “A life worth living”. Animals, 6(6), pp. 2–7. doi: 10.3390/ani6060035.

Wiese, T. R., Haskell, M., Jarvis, S. et al. (2023) Concerns and research priorities for Scottish farmed salmon welfare – An industry perspective. Aquaculture, 566, p. 739235. doi: 10.1016/j.aquaculture.2023.739235.

Yang, L. et al. (2021) Computer vision models in intelligent aquaculture with emphasis on fish detection and behavior analysis: a review. Archives of Computational Methods in Engineering. Springer Netherlands, 28(4), pp. 2785–2816. doi: 10.1007/s11831-020-09486-2.

CHAPTER 4. Application of Qualitative Behavioural Assessment (QBA) as a welfare indicator for farmed Atlantic salmon (*Salmo salar*) in response to a stressful challenge

Timothy Robert Wiese[1], Sonia Rey Planellas[1], Marie Haskell[2], Susan Jarvis[3], Andrew Davies[4], Francoise Wemelsfelder[2], James F Turnbull[1]

[1]: Institute of Aquaculture, University of Stirling, Stirling, FK9 4LA, UK.
[2]: SRUC (Scotland's Rural College),West Mains Road, Edinburgh, EH9 3JG, UK
[3]: Global Academy of Agriculture and Food Systems, University of Edinburgh, Midlothian, EH25 9RG, UK
[4]: Aquascot Ltd., Fyrish Way, Alness, Ross-shire, IV17 0PJ, UK.

4.1 Abstract

Animal welfare assessments have historically struggled to find a balance between investigating the emotional states of animals and upholding objectivity by focusing solely on the empirical evidence available. Behavioural analysis may provide one of the few insights into the more subjective experiences of an animal without compromising the scientific approach. One example of such a tool is Qualitative Behavioural Assessment, or QBA. Rather than assessing a set of separate physical behaviours (i.e., what is the animal doing, such as swimming, biting or feeding), QBA specifically focuses on the overall manner in which an animal executes its behaviour (i.e., how it is behaving, such as relaxed or stressed). Through its integrative approach, QBA enables a 'whole-animal' assessment of how an animal expresses these qualities through its behaviours (i.e., its expressive characteristics). QBA has been validated for a range of terrestrial farmed animals, offering a time-efficient and non-intrusive approach that yields unique insights for welfare assessments. However, the application of QBA in aquaculture remains largely unexplored, and no studies have yet examined QBA's ability to capture the welfare of fish that has been compromised following exposure to stressful events. With increasing scientific evidence and public opinion that fish are sentient, there is a growing demand for the development and implementation of tools that are suitable for assessing the emotional states and welfare of farmed fish. This study therefore aimed to investigate the use of QBA in Atlantic salmon, assessing its ability to capture changes in their emotional state after exposure to stressful events.

For this study, 9 tanks of juvenile Atlantic salmon were video-recorded every morning for 15 minutes, over a 7-day period, in the middle of which a stressful challenge (i.e., an intrusive sampling event) was conducted on the salmon. Each video clip was recorded when lights were first switched on in the mornings, and later edited to include the first full minute for which the salmon were in clear view after the lights had turned on. The resultant 63 video clips were then semi-randomised to avoid predictability and treatment bias from scorers before being used within the QBA. The initial stage of QBA first had 12 observers collectively generate a list of 16 qualitative descriptors (or terms, e.g., relaxed, agitated, stressed), after viewing unrelated video recordings depicting a range of expressive characteristics from salmon in different contexts. In the second stage, a different group of 5 observers, who were blind to the treatment (i.e., pre- or post-stressful challenge) and had varied experience with salmon farming, subsequently watched the 63 video clips, and scored the 16 qualitative terms for each clip using a Visual Analogue Scale (VAS). The QBA scores from the 5 observers were analysed together using Principal Components Analysis (PCA, correlation matrix, no rotation) to identify perceived patterns of expressive characteristics across the video clips. The PCA revealed 4 dimensions that collectively accounted for 74.5% of the variation between video clips, with PC1 (relaxed / content / positive active vs. unsettled / stressed / spooked / skittish / agitated) explaining the highest percentage of variation (37%). Scores for video clips on PC1, PC2, and PC4 achieved good inter- and intra-observer reliability. There was a significant difference between PC1 scores before and after the salmon were exposed to the challenge ($p = 0.03$), indicating that the salmon were perceived as more stressed after the sampling had taken place. In addition, PC1 scores were positively correlated to the degree and frequency of darting behaviours recorded from 2 separate sets of video clips (same clips as QBA: $r = 0.42$, $p < 0.001$; 1-minute post QBA clips: $r = 0.33$, $p < 0.01$). The results from this study are the first to validate QBA's sensitivity to changes in the expressive characteristics of Atlantic salmon following exposure to putatively stressful events, and further demonstrate QBA's potential as a welfare indicator within aquaculture.

Keywords:

Emotional state; aquaculture; positive welfare; behavioural analysis; qualitative behaviour assessment

4.2 Introduction

Animal welfare science has faced the challenge of addressing all aspects of welfare without compromising objectivity and the need for empirical evidence. Physical health has long been recognised as an essential component of animal welfare (Fraser *et al.*, 1997; Dawkins, 2004; Franks, Ewell and Jacquet, 2021). However, a widely held perspective now is that animal welfare is ultimately a state that is perceived by the animal itself, and we should therefore also include concerns for the animal's mental well-being (Green and Mellor, 2011; Hemsworth *et al.*, 2015; Fife-Cook and Franks, 2019). There is thus a growing demand that welfare assessments, including those for fish, adopt a more holistic approach that places additional focus on monitoring the animal's positive experiences (Boissy *et al.*, 2007; Mellor, 2016; Fife-Cook and Franks, 2019; Rault *et al.*, 2020; Franks, Ewell and Jacquet, 2021; Veit and Browning, 2021; Browning, 2023). Welfare appraisals that adopt this integrated approach, however, inevitably enter the murky waters that are an animal's subjective experiences (Dawkins, 2003, 2015). Despite decades of research trying to resolve this issue, the only progress thus far has been reaching a consensus that there is no single "measure" that can adequately cover what welfare entails (Mason and Mendl, 1993; Broom, 1998; Dawkins, 2003; Stien *et al.*, 2013). This dilemma has resulted in the mental well-being of fish often being overlooked in welfare assessments (Jarvis *et al.*, 2021).

In 2018, Atlantic salmon accounted for 4.5% of global aquaculture production by tonnage (FAO, 2020). In 2021, production of Scottish Atlantic salmon reached an all-time high of 205,393 tonnes, with more than 50 million smolts transferred to sea in the same year (Munro, 2022). Total tonnage of Scottish farmed salmon, relative to the number of employees on-site, has increased 11-fold within seawater and 6-fold within freshwater between 1985-2016 (Ellis *et al.*, 2016). This increase in the numbers of fish relative to farm staff, unavoidably reduces the time available for monitoring the salmon. There is also mounting scientific evidence supporting the sentience of fish (Sneddon, Braithwaite and Gentle, 2003a; Chervova and Lapshin, 2011; Brown, 2015; Kristiansen *et al.*, 2020). A UK National survey, involving 1963 members of the public, found that 77% agreed or strongly agreed that fish can feel pain, and 80% agreed that this should therefore be of concern (Rethink Priorities, 2019). Considering the scale of this industry, there is a clear ethical and economic incentive to develop welfare indicators that are not only practical, but also attempt to include aspects of mental well-being (both positive and negative) in their assessment.

To achieve such an assessment, a framework was proposed in which welfare assessments are viewed in the context of a simple question: "Is the animal healthy, and does it have what it wants?" (Dawkins, 2003). Considering that what animals want may not always be what is best for their welfare (Veit and Browning, 2021), this question has been rephrased to: "Provided an animal's health is given precedence, are its desires or preferences being met?". Answering the second, difficult part of this question (i.e., delving into an animal's subjective experiences) may require accepting two arguments. Firstly, that consciousness still presents an impasse for scientific study (Fraser *et al.*, 1997). Secondly, given that animals cannot express their desires and needs in human language, behavioural analysis may provide some of the best insights into what they "want" (Dawkins, 2015). Behaviours exhibited by an animal are, in essence, the final product of all its own decision-making processes (Dawkins, 2004; Mendl, Burman and Paul, 2010). They are the "final common path", as described by Sherrington (1906), or the "ultimate phenotype" and "expression of the emotions" (Darwin, 1872). Behavioural analysis provides a number of additional advantages over physiological and morphological measures in welfare assessments. Such analyses are frequently non-intrusive (the animal is unaware it is being assessed), and are often quick to observe (Dawkins, 2004; Huntingford *et al.*, 2006; Martins *et al.*, 2012). Behaviour is also gaining recognition as a general, pre-clinical 'early warning system' for issues that may be emerging within the stock (Dawkins, 2004; Huntingford *et al.*, 2006; Oppedal, Dempster and Stien, 2011; Duthie *et al.*, 2020; Wiese *et al.*, 2023).

Moreover, through collaborative development of behavioural assessment tools with farmers, it may be possible to externalise the tacit knowledge farmers tend to have of the well-being of their animals (Hoffmann, Probst and Christinck, 2007). Such knowledge, typically gained through years of experience, enables farmers to detect more subtle changes in an animal's behaviour when issues arise, even though the exact nature of the problem may remain unclear (Hoffmann, Probst and Christinck, 2007). Qualitative Behavioural Assessment (QBA) is a behavioural assessment tool that benefits from this approach while eliminating the associated drawbacks relating to reproducibility and validity (Browning, 2022). QBA is an integrative assessment of the "whole-animal", where observations are made on the animal's body language (including their appearance, behaviour, and interaction with others and the surrounding environment) as an indicator of its welfare state (Wemelsfelder, 2007; Ellingsen *et al.*, 2014; Cooper and Wemelsfelder, 2020; Vasdal *et al.*, 2022). Different aspects of the animal's expressive characteristics are summarised through a number of 'descriptors' (or terms) such as: calm, inquisitive, agitated, or stressed (Wemelsfelder *et al.*, 2001; Jarvis *et al.*, 2021; Vasdal *et al.*, 2022). By summarising such various expressive characteristics, QBA focuses less on what an animal does, and more on how it does it (Wemelsfelder *et al.*, 2001). These terms are used with the intention of covering the full range of both negative and positive emotions (Jarvis *et al.*, 2021; Browning, 2023).

Previous studies have validated the use of QBA against other welfare indicators for various livestock species, and demonstrated high degrees of inter-observer reliability between observers (Fleming *et al.*, 2016; Minero *et al.*, 2018). Additionally, QBA allows for simple, time-efficient, and non-intrusive assessments of an animal's well-being (Ellingsen *et al.*, 2014; Browning, 2022). QBA is also the only measure currently included in the EU Welfare Quality® welfare assessment protocols to assess positive emotional states in cattle, pigs, and poultry (Welfare Quality®, 2009; Keeling *et al.*, 2013). To date, however, the only QBA study to be applied to fish examined solely the inter- and intra-observer reliability and QBA's association with ethograms of salmon behaviour, without the inclusion of any treatments (Jarvis *et al.*, 2021). No studies have yet examined fish exposed to stressors, or compared QBA scores in this context to other welfare indicators. Comparing QBA scores against other welfare indicators for salmon may help to further explore what potential role QBA may have as a welfare assessment tool. Darting represents a behavioural response previously recorded in fear-conditioning studies of fish, and is commonly associated with predator avoidance (Magurran and Pitcher, 1983; Cantalupo, Bisazza and Vallortigara, 1995; Domenici and Blake, 1997; Ashley and Sneddon, 2008). It is considered a stress response which, when increasing in frequency or intensity, may indicate impaired welfare (Magurran and Pitcher, 1983; Ashley and Sneddon, 2008; Nomura *et al.*, 2009). Feed intake is also generally considered a reliable indicator within health and welfare assessments of farmed fish (Jobling *et al.*, 2001). A loss in appetite is potentially a sign of impaired welfare (Schreck, Olla and Davis, 1997; Huntingford *et al.*, 2006). The main aim of this study was therefore to examine QBA's ability to detect differences in the expressive characteristics of Atlantic salmon after exposure to a stressful challenge (i.e., an intrusive sampling event). In addition, this study also aimed to compare these QBA scores against other welfare indicators for salmon; their daily feed intake (as a proxy for appetite) and darting behaviours (i.e., sudden, rapid movements of the salmon).

4.3 Materials and methods

4.3.1 Ethical review

Ethical approval for the recording of salmon and QBA work was obtained from the University of Stirling's Animal Welfare & Ethical Review Body (Approval reference no. 2022-6783-5196).

4.3.2 Experimental set-up

4.3.2.1 Animals

The juvenile Atlantic salmon used in this study were transferred on November 16th, 2021, from the Niall Bromage Freshwater Research Unit (NBFRU), Denny, to the Marine Environmental Research Laboratory (MERL) in Campbeltown, Argyll and Bute, Scotland. The salmon were around 14 months of age, and weighed on average 285-360 grams. There were ~80 smolts in each tank at the start of the recording, with an average stocking density of ~34kg/m^3.

4.3.2.2 Husbandry

The salmon were housed in a total of 9 identical flow-through tanks (1.4m diameter, 750L volume). Seawater was filtered through a Lacron sand filter (4x100 micron bag filters) before flowing into the tanks to minimise turbidity. Automatic belt feeders provided standard salmon pelleted dry feed (Skretting Nutra Advance / Supreme©) to all tanks every 20 minutes between 05:00-09:00 and 16:30-23:30. Dirty water and uneaten feed were flushed out of the tanks through standpipes daily, between 09:00-09:15. Any mortalities found during this period were immediately removed. Lights were turned on at exactly 10:30am each morning.

4.3.2.3 Treatments (including stressful challenge)

Video clips for this study were recorded around a stressful challenge, conducted on February 18th, 2022. This stressful challenge involved a sampling event which was carried out for another study on these salmon. This required capturing, anaesthetising, and handling each of the salmon out of water for measuring their weight, length, and condition factor. While feed withdrawal was also required 24 hours before sampling could be carried out, the recording schedule was designed on the assumption that the main disturbances (i.e., stressful challenge) to the salmon would occur largely as a result of this sampling event. For the purposes of the study that involved the sampling event, a subset of the salmon that were sampled were then euthanised in accordance with schedule 1 protocols in order to obtain their hepatosomatic index. Following the sampling event, there were approximately 50 salmon left in each tank, with an average stocking density of ~21kg/m^3 (Figure 4-1).

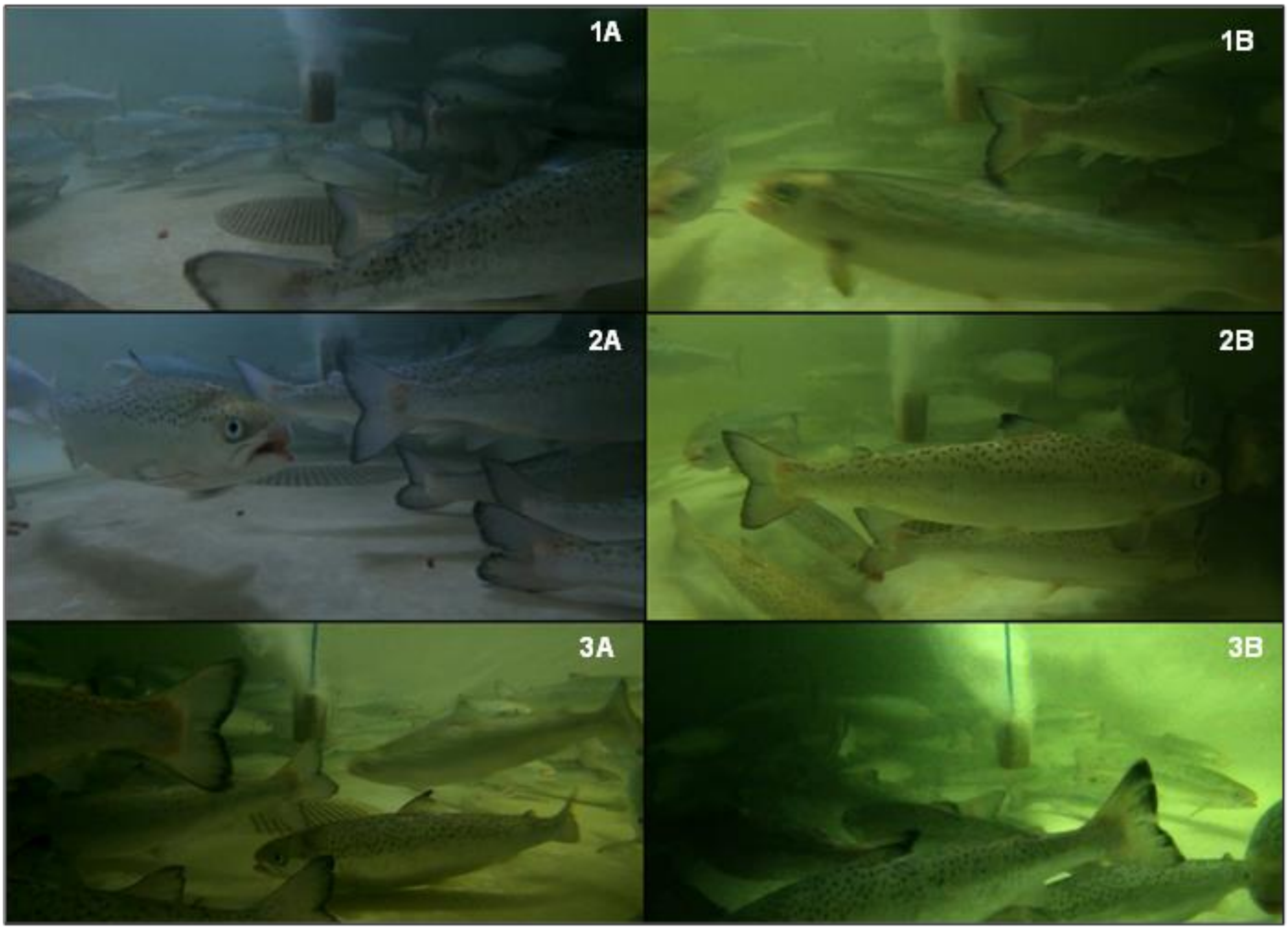

Figure 4-1. Screenshots comparing views of the same tanks before (1A-3A) and after (1B-3B) the sampling event and consequent reduction in stocking density. Snapshots taken from tanks 1, 3, and 5 on the first baseline day (A) and the first day post-sampling (B).

4.3.2.4 Camera and tanks set-up

Cameras were installed in the tanks to record video clips for the QBA and behavioural assessments. To do this, every morning at 9am, GoPro Hero9 Black© cameras were installed at 1m depth using a fixed metal pole, which was positioned flush against the inside of each tank to ensure the same angle and field of view (FOV) for recordings. This was carried out 90 minutes before lights went on to allow time for salmon to acclimatise to the cameras. These cameras were also installed each morning for 2 days before recording commenced to allow the salmon to further acclimatise to these novel objects. To minimise any additional disturbances, cameras were turned on before being submerged with recording controlled remotely through the GoPro Quik© mobile application. Connectivity from mobile phone to each underwater camera was achieved through the use of coaxial cables taped to each device. Coaxial cables conduct electrical signals (including Wi-Fi) through an insulated shield, extending network connections to a submerged device (e.g., camera). Recordings for each tank were taken on a strict daily schedule, after lights went on, to ensure consistency. A minimum of 15 minutes were recorded for each tank once lights went on. All personnel on-site strictly avoided carrying out any procedures around the tanks during filming.

4.3.2.5 Recording schedule

A 7-day period of video recording was scheduled to gather footage for all behavioural analysis (i.e., QBA and darting behaviours), with the stressful challenge (i.e., sampling) conducted during the middle of this period. Sampling was carried out on all 9 tanks of salmon on February 18th, 2022. To obtain a ‘baseline’ and account for any potential day to day variation in behaviour, 3 consecutive days were recorded before the stressful challenge occurred. A further 3 consecutive ‘post-sampling’ days were required for recording the salmon’s recovery from this stressful challenge. Figure 4-2 provides a summary of the recording schedule. The ability for QBA to reflect any impacts on salmon behavioural

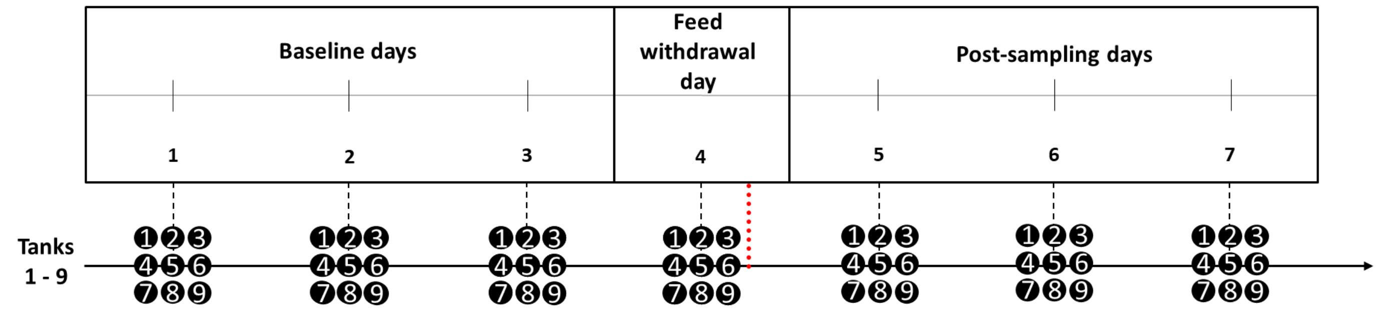

Figure 4-2. Recording schedule and timeline for experiment. Black dots represent each time a tank was recorded for the day, and the dashed red line (after day 4) illustrates when the stressful challenge (sampling event) occurred.

expressions, as a result of these disturbances, could then be assessed from these recordings (section 2.3.1.1 outlines how the video clips were prepared for QBA).

4.3.3 Qualitative Behavioural Assessment (QBA)

The QBA process consisted of two main stages. Stage 1 involved 12 observers in the generation of the QBA terms for describing the salmon's expressive characteristics and stage 2 involved 5 different observers scoring the QBA terms for each of the video clips.

4.3.3.1 Stage 1 – term generation

Twelve professionals employed in the Scottish salmon farming industry were recruited for the term generation stage, which involved two separate meetings. All participants had at least one year of experience working directly with farmed salmon, with a number of participants in senior / management roles. During term generation, various video clips were used which were taken from different farm sites under different contexts (e.g., during the middle of the day or during feeding, after treatments / transportation etc.). In this study, we define 'expressive characteristics' as the extent to which qualitative characteristics of salmon behaviour (e.g., relaxed, purposeful, lethargic, stressed) are expressed. The video clips were selected to represent all four aspects, or 'quadrants', of behavioural expression (high to low energy, positive to negative valence) as outlined by Mendl, Burman and Paul, 2010 (see Figure 4-3).

Before terms were generated by participants, the theory and practice of QBA was explained to them and they were provided with guidance on how to generate appropriate terms. To avoid bias from instructors, examples of classic terms from terrestrial farming systems were used. After the first meeting, participants were asked to individually watch the video clips in advance of the second meeting and generate their own personal list of terms. During the second meeting the participants discussed these terms, including how they should be divided between the 4 quadrants of behavioural expression (high to low energy, positive to negative valence). Participants were then asked to select a maximum of 20 terms which were balanced across the 4 quadrants, and best described the range of salmon behavioural expression. By the end of the meeting, the group had agreed on 16 terms. These included

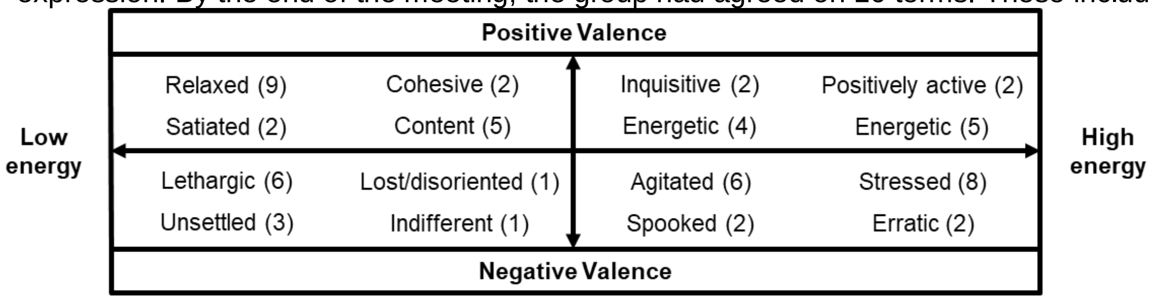

Figure 4-3. Final list of Qualitative Behavioural Assessment terms generated from stage 1. Valence (positive / negative) and energy (high / low) were used to help describe and discuss terms across the 4 quadrants. Numbers in brackets indicate the total number of participants who brought each term to the initial meeting.

the terms “diving deep” and “flighty”, which were excluded by the experimenters from the final list used in the second stage. QBA requires terms that convey some aspect of emotional state and the term “deep diving” did not. Other terms (e.g., spooked, erratic, unsettled, agitated) already covered aspects of the term “flighty”. The final QBA term list therefore had 16 terms, with 4 in each quadrant of behavioural expression (Figure 4-3). These terms were then used in the QBA scoring stage.

4.3.3.2 Video preparation before stage 2

For use in the QBA scoring stage, shorter video clips were extracted from each of the 63 15-minute videos. These clips were the first full minute that the salmon remained clearly in view, starting from 30 seconds after the lights were turned on. This excluded the initial “noise” from the salmon’s startle responses to the lights. Video clips were first randomised with respect to their chronological order and their occurrence before or after the sampling order. To facilitate observer concentration and motivation, they were then arranged so that clips showing contrasting expressive characteristics (e.g., primarily high energy, negative valence vs. low energy, positive valence) were distributed evenly throughout the scoring sessions. Unknown to observers, 4 of the original 63 video clips were duplicated to allow for an assessment of intra-observer reliability (the degree to which participants showed agreement within their own scoring sessions). This resulted in a total of 67 video clips being scored by each observer.

4.3.3.3 Stage 2 – QBA training and scoring session

Scoring sessions for the QBA were carried out with a new group of five observers. These five observers consisted of two Post-doctoral fish welfare researchers from the University of Stirling, and three industry professionals all with a higher degree education and between 3-20 years of aquaculture industry experience. These observers consequently had a varied level of experience in working with and observing salmon. All but one observer had hands-on experience in salmon husbandry in a commercial setting.

Observers were given online training in QBA. A brief introduction was given on the principles of QBA and the general purpose of this study (i.e., exploring the use of QBA within fish). Observers were kept blind to treatment (i.e., the stressful sampling challenge), and were instead only informed about the general context behind the video clips (location of filming, number of tanks and days involved in the recording). To ensure everyone’s understanding of the terms was aligned, an open discussion was first conducted. The meaning behind the terms was explored, and observers were invited to raise any questions about terms which required clarification. General instructions were given on how to assess whole animal expressivity and how to use the Visual Analogue Scales (VAS) to score the prevalence of each term within a video clip. A VAS is a measurement instrument that allows for the scoring of characteristics (such as those of behavioural expressions) that are believed to range across a continuum of values (Gould, 2001). Observers were reminded that terms must be scored independently from each other, so that in situations where there were contrasting expressive characteristics among different salmon (e.g., some appearing agitated and others relaxed), those contrasting terms could both receive high scores for the same clip.

All QBA scoring was carried out on scoring sheets developed on SurveyMonkey®. For each term, a horizontal line with a 100-step scale was presented as a VAS, along which a single mark could be made. The distance from the left end of the scale would correspond to the participant's assessment of the intensity for each term observed. The left end of the scale represented complete absence of an expressive characteristic described by a term, whereas the right end represented the maximum expression for the term (e.g., the salmon could not be more erratic). To minimise any potential influence on scoring, no quantitative values would appear alongside the 1-100 step VASs as observers carried out the QBA. They were encouraged to use the entire scale when judging the intensity of each expressive characteristic. Video clips were labelled according to their order in the scoring sheets and transferred electronically to the group. Due to the large number of clips, observers were instructed to avoid scoring them all in a single session, but also to carry out their scoring sessions with minimal delay between each other (i.e., within the same week) to minimise potential variation introduced by scoring on different days.

4.3.4 Additional welfare measures – feed intake and darting events

4.3.4.1 Feed intake

Feed input and feed waste were recorded for each tank daily alongside the 7 days of QBA recordings. Feed intake was then determined by subtracting feed waste from feed input, and analysed at a 'per individual' value. After the sampling event, the amount of feed supplied was adjusted to the biomass of salmon remaining in the tanks.

4.3.4.2 Darting behaviour

For the purpose of this study, darting behaviours were defined as a "rapid, burst of movement clearly distinct from the salmons' regular swimming behaviours; this includes sudden changes in direction, acceleration, and/or positioning of the salmon in the tank". A method was created to record 'darting events' in the same 63, 1-minute video clips used for the QBA. Since any of these darting events would have also been observable during the QBA, and thus potentially affected the scoring of certain QBA terms, another second set of video clips were also investigated. This second, separate set involved an additional 63, 1-minute video clips that were taken immediately after the QBA clips.

To allow multiple darting events to be recorded in one clip, any darting behaviour must have stopped before the next event could be recorded. The number of salmon involved in each darting event was first recorded and categorised by the proportion to the total number of salmon in the tank (Table 4-1). Weighted scores were then assigned to each of these categories, relative to their proportions (Table 4-1). This was done to provide additional granularity with respect to the magnitude of darting involved. A final score was then calculated for each clip, based on the sum of weighted scores from all darting events recorded.

Table 4-1. Categories of darting events by the proportion of salmon from the tank involved, as well as their corresponding weighted scores.

Proportion of salmon in tank involved in each darting event:	Weighted score:
Less than 4%	1
Less than 8%	2
Less than 15%	3
More than 15%	4

Video playback speed was altered to ensure the number of salmon involved were counted correctly. Where the number of salmon darting was too high to allow for counting, the event was then categorised as involving more than 15% of the fish in the tank.

4.3.5 Statistical analyses

4.3.5.1 Data handling of QBA scores

For each QBA score, the distance of each observers' marks from the zero point of the scales was automatically measured and recorded by SurveyMonkey. The complete dataset of these raw QBA scores were then imported from SurveyMonkey into Microsoft Excel (Version 2301). Data was organised into a matrix, with QBA terms listed horizontally in the first row and video clip numbers and labels in the first few columns. Unless otherwise stated, all statistical analyses were run in R Studio (version 4.2.2). The threshold of significance for any statistical test was $p < 0.05$. For the Linear Mixed Effects Model (LMEM) analyses conducted later in the study, the package "nlme" was applied.

4.3.5.2 Principal Component Analysis (PCA)

A Principal Component Analysis (PCA) was carried out, using a correlation matrix on the entire dataset of QBA scores to reduce the dimensionality of the QBA terms. This specific form of PCA was chosen because, while the 16 different terms used the same 100-step VAS in scoring, it is more likely that observers would use these scales differently depending on the term involved. Consequently, the different terms / variables cannot technically be assumed to be scored on the same scale. PCA allows for the 16 terms scored within each video clip to be summarised by a numerical value for each Principal Component (i.e., the PC "score"). No post-processing step of 'rotation' was carried out, as the only goal of the PCA was to reduce the dimensionality of terms.

The highest positively and negatively loaded terms for each component were identified which, together, represented the larger pattern of expressive characteristics illustrated within each PC. To determine whether PCs were eligible for further analysis, a combination of criteria was used. Following the "Kaiser criterion", which states that the number of factors to retain should correspond to the number of eigenvalues greater than one, only PCs with eigenvalues >1 were considered (Kaiser, 1960). Within each component, there also had to be good inter-observer reliability in the PC scores (section 2.5.3). There also needed to be a coherent biological interpretation of the terms that had the highest positive and negative loadings within each component. For example, a higher score for PC1 suggested that salmon were more unsettled / stressed, whereas a lower score suggested that salmon were more relaxed / content.

For the complete set of PC scores obtained, Q-Q plots, histogram symmetry, skewness and kurtosis values, sphericity, and Leven's test were inspected to ensure all assumptions required for carrying out further parametric tests were met (including normality of data). The scree plot and proportion of variance for each PC were also used as additional guidance for determining the inclusion of PCs in further analysis.

4.3.5.3 Inter/Intra-observer reliability

Kendall's coefficient of concordance (W) was used to calculate the level of agreement between the 5 participants' PC scores in the combined data set, for each of the PCs. Any value of W less than 0.4 was considered to reflect unacceptable inter-observer variability. This analysis was carried out using IBM SPSS Statistics 28 (IBM Corp., 2021). The degree to which observers showed agreement between their scores of the duplicated video clips was (given normal distribution of the scores) determined using Pearson's correlation, performed on each of the relevant PC scores.

4.3.5.4 Comparing Pre vs. Post disturbances

QBA scores of the salmon before and after the stressful challenge were analysed by applying separate Linear Mixed Effects Models (LMEM) to each of the relevant PCs (PC1, PC2, and PC4). For each LMEM, the PC score was the dependent variable, 'Pre vs. post disturbance' and 'Observer' were fixed factors, and tank number was a random factor. Before the LMEMs were applied, ANCOVAs were first carried out (with day number as a covariate) to ensure that there were no significant time trends within each subset of days 1-3 and 5-7. Since no additional time trends were present within these subset of days, day number was also included in the LMEMs as a random factor.

Although Kendall's coefficient determines whether there is good agreement between observers for PC1, PC2, and PC4, the actual "treatment" effect of observers still needed to be accounted for, hence the inclusion of 'Observer' as a fixed factor.

4.3.5.5 Comparing feed intake and darting events with QBA scores

For each tank every day, feed intake and two separate sets of darting scores were recorded (2 x separate sets of 63 video clips). Similar LMEMs were applied, with tank and day number as random factors, to first determine whether 'Pre vs. post disturbance' had a significant impact on each of these additional measures. Spearman correlation tests were then carried out to compare feed intake and the two separate sets of darting scores against the corresponding mean PC scores of the 63 clips used in the QBA. Mean PC scores were derived by averaging the PC scores from the 5 observers. Correlations were only carried out against principal components with scores that were significantly different between 'Pre vs. post disturbance'.

4.4 Results

4.4.1 Qualitative Behavioural Assessment

4.4.1.1 Principal Component Analysis

PC1, PC2, PC3, and PC4 had eigen values > 1. PC1 explained the greatest percentage of variation at 37%, with the first four components collectively explaining 74.5% of the variation in the data (Table 4-2).

Table 4-2. Eigen analysis of principal component 1, 2, 3, and 4.

Value	PC1	PC2	PC3	PC4
Eigen value	5.88	2.82	1.95	1.27
% of variation explained	36.7%	17.7%	12.2%	7.9%
Cumulative %	36.7%	54.4%	66.6%	74.5%

As outlined in Table 4-3, PC1 ranged from relaxed / content / positive active to unsettled / stressed / spooked / skittish / agitated. For PC2, the only positively loading term was relaxed, with the main negatively loading terms being energetic / purposeful / inquisitive.

Table 4-3. Qualitative Behavioural Assessment term loading values for each principal component. The highest negatively and positively loaded terms for each PC are in bold.

Term	PC1	PC2	PC3	PC4
Relaxed	**-0.359**	**0.074**	-0.003	0.073
Agitated	**0.309**	-0.272	-0.090	-0.109

Inquisitive	-0.197	**-0.366**	-0.151	0.185
Unsettled	**0.358**	-0.185	-0.073	-0.049
Cohesive	0.039	-0.153	0.297	**-0.491**
Spooked/Skittish	**0.327**	-0.199	-0.024	-0.112
Positive active	**-0.286**	-0.286	**-0.168**	0.110
Indifferent	-0.148	-0.254	**0.477**	-0.067
Purposeful	-0.145	**-0.395**	**-0.284**	-0.104
Erratic	0.226	-0.226	-0.038	**0.431**
Energetic	-0.156	**-0.459**	**-0.202**	-0.029
Lost/Disoriented	0.103	-0.165	**0.356**	**0.585**
Satiated	-0.224	-0.186	0.232	**-0.290**
Lethargic	0.025	-0.178	**0.560**	0.066
Stressed	**0.332**	-0.171	-0.056	-0.211
Content	**-0.358**	-0.042	0.009	-0.050

Figure 4-4 illustrates the relationship that the QBA terms have with both PC1 and PC2. For example, a more negative PC1 score indicates salmon that were more relaxed, content, and positive active. PC1, PC2, and PC4 demonstrated acceptable inter-observer reliability for their PC scores (PC1: W = 0.63, X^2 = 207.57, p < 0.001; PC2: W = 0.46, X^2 = 152.19, p < 0.001; PC4: W = 0.56, X^2 = 184.94, p < 0.001). All four PCs showed acceptable intra-observer reliability between PC scores of video clips that were duplicated (PC1: *r* = 0.716, p<0.001; PC2: *r* = 0.755, p<0.001; PC3: *r* = 0.552, p<0.05; PC4: *r* = 0.581, p<0.01). PC3 had a W value below 0.4, which was considered unacceptable and therefore not included in further analysis. PC1, PC2, and PC4 were retained for further analysis.

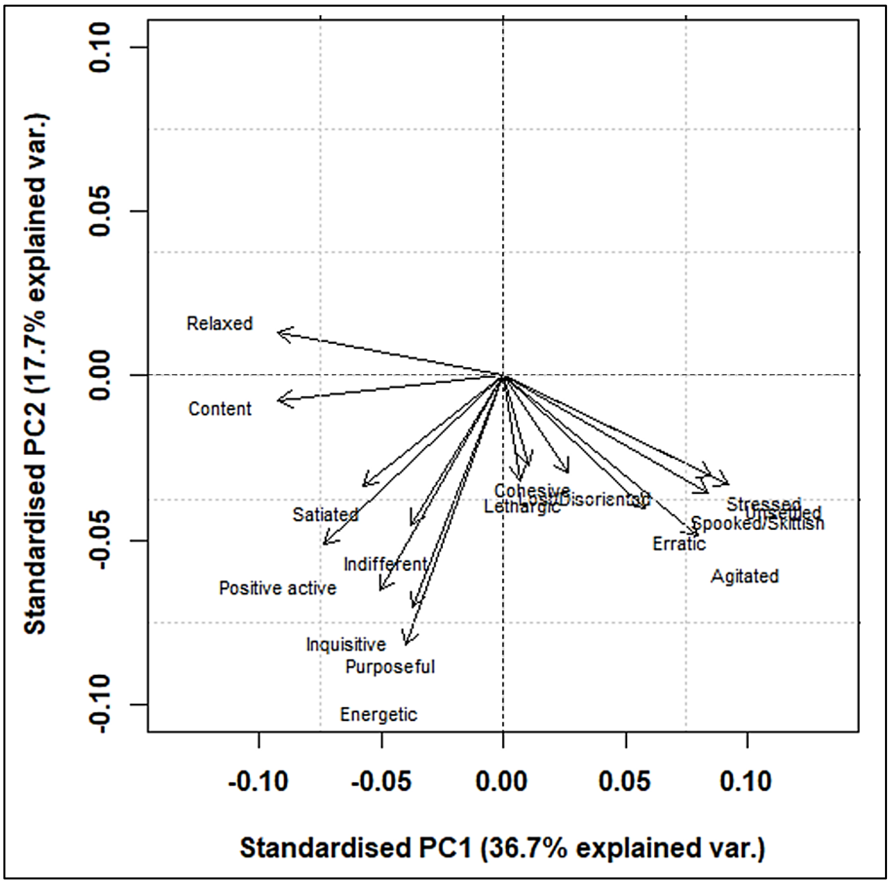

Figure 4-4. Loading plot of all 16 Qualitative Behavioural Assessment (QBA) terms used in this study for PC1 and PC2. Axes represent standardised eigen vectors for which the QBA terms load onto the two main principal components of the analysis. A more negative value for PC1 indicates an overall higher score for relaxed, content, and positive active.

4.4.1.2 Effect of the stressful challenge (intrusive sampling) on PC scores

There was a significant difference between PC1 scores when comparing days before and after the stressful challenge (p = 0.03, Figure 4-5). PC1 scores (averaged between the 5 observers for each video clip) ranged from -4.97 to 6.04. The mean difference between PC1 scores for pre vs. post-disturbance days was + 0.82 (Pre = -0.239, Post = 0.584). Overall, all five observers scored PC1 higher for post-disturbance days. 7 out of 9 tanks received a higher average PC1 score for post-disturbance days. Figure 4-6 illustrates the comparative likelihood of a PC1 score being higher or lower for video clips that were recorded either before or after the sampling event. No significant differences were found for PC2 and PC4 scores ($p > 0.05$). For PC1, PC2, and PC4, there was a significant effect for observers as a fixed effect ($p < 0.001$).

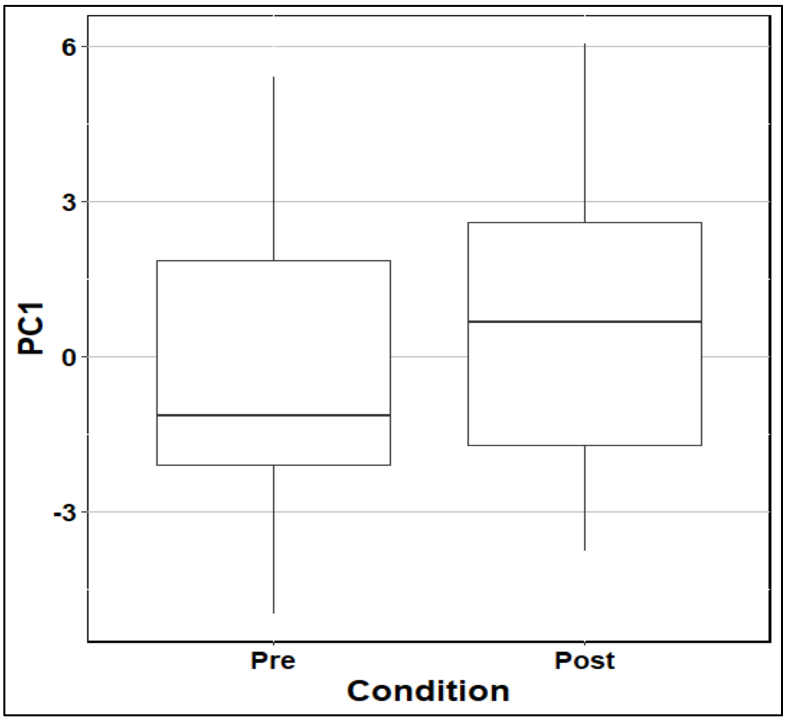

Figure 4-5. Box plot to compare differences in spread of PC1 scores before and after feed withdrawal and sampling events.

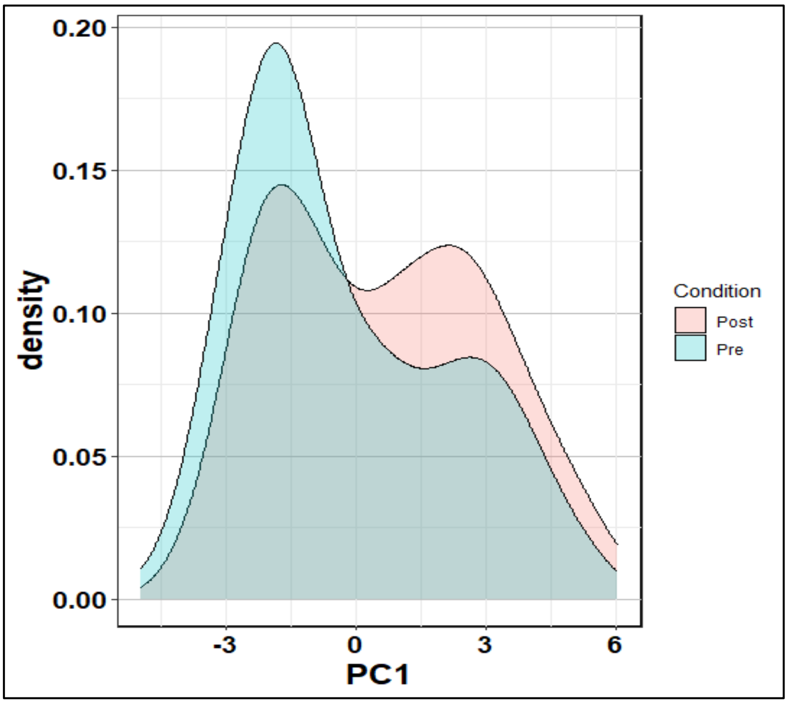

Figure 4-6. Layered density plot comparing different probabilities of various PC1 scores occurring, depending on whether they were taken pre vs. post disturbance.

4.4.2 Feed intake, darting behaviours, and their association with QBA

A significant difference was found in the feed intake of salmon from tanks before and after the stressful challenge ($p = 0.002$). Mean daily feed intakes per fish were 2.11g for pre-disturbance (SEM = 0.06) and 1.58g for post-disturbance (SEM = 0.15), resulting in an average 0.53g reduction in daily feed intake per fish post-disturbance. However, there was no significant association found between mean PC1 scores and feed intake ($r = -0.10$, $p > 0.05$). No significant difference was found between darting scores before and after the stressful challenge, in either set of 63 video clips used (same clips as QBA: $p > 0.05$; 1-minute post QBA clips: $p > 0.05$). However, PC1 scores showed a moderate positive correlation with the darting scores taken from either set of video clips (same clips as QBA: $r = 0.42$, $p < 0.001$; 1-minute post QBA clips: $r = 0.33$, $p < 0.01$, see Figure 4-7).

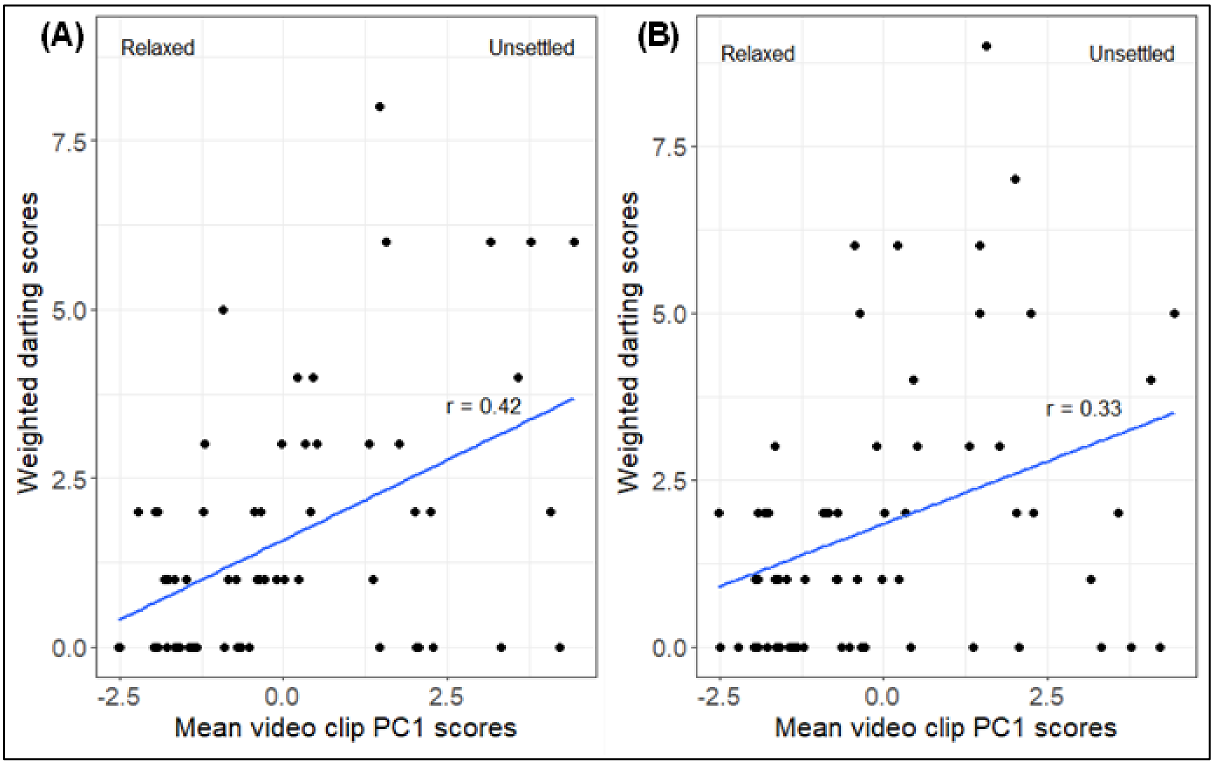

Figure 4-7. Scatterplot of mean PC1 scores (Relaxed – Unsettled) for video clips vs. **(A)** weighted darting scores calculated from the same clips used for the QBA, and **(B)** weighted darting scores calculated from video clips taken one-minute after QBA clips. Line of best fit and r correlation coefficients from spearman correlation tests included.

4.5 Discussion

Integrating indicators of the emotional state of animals within welfare assessments has previously proven to be problematic for many reasons. This study’s aim was to determine QBA’s ability to detect the effects of a stressful event on Atlantic salmon. We applied QBA to quantify and evaluate the expressive characteristics of Atlantic salmon before and after exposure to a putatively stressful challenge. While feed withdrawal was required before sampling could be carried out, the sampling event was the focal point as the experimental treatment of this study. The process of capturing, anaesthetising, and handling salmon out of water for sampling has been described as intrusive, stressful, and detrimental for welfare (Djordjevic *et al.*, 2012; Zahl, Samuelsen and Kiessling, 2012; Santurtun, Broom and Phillips, 2018)**.** Previous studies that have assessed how salmon recover from stressful events (e.g., handling / anaesthesia / invasive sampling) often monitored the recovery over a 24-72hr period (Iversen, Finstad and Nilssen, 1998; Sandodden, Finstad and Iversen, 2001; Djordjevic *et al.*, 2012). Thus, a 3-day period for both the baseline and 'recovery' stage was considered to be sufficient for the purpose of this study.

There was acceptable agreement between the five observers in this study, who were blind to the treatment and had varied experience in monitoring fish behaviour / welfare. There was one main dimension of QBA that proved effective in capturing changes in the emotional state of the salmon within this study; relaxed / content / positive active – unsettled / stressed / spooked / skittish / agitated (PC1). PC1 explained the largest proportion of variation in expressive characteristics of the salmon (36.7%). There were significant differences between PC1 scores before and after the stressful challenge (sampling), with salmon being scored as more unsettled / stressed / spooked / skittish / agitated after sampling. This reflected a shift from low energy, positive valence to high energy, negative valence after sampling, a contrast that was consistently recorded by all observers and in the majority of tanks. In addition, the single recording that was perceived the most "positively" (i.e., the most relaxed / content / positive active) was taken before any potential impacts from sampling had occurred. These results are in agreement with numerous papers that have previously used QBA to assess the emotional state of terrestrial farmed animals under similar challenging interventions (e.g., for cattle, horses, pigs, and hens), with PC1 typically being characterised by terms such as relaxed and content vs. agitated (Napolitano, Rosa and Grasso, 2012; Rutherford *et al.*, 2012; Fleming *et al.*, 2013; Sant'Anna and Paranhos da Costa, 2013; Muri *et al.*, 2019). Furthermore, these past studies have used similar descriptors to describe the other main terms used in PC1 for this study; unsettled (uneasy), stressed (nervous), spooked / skittish (scared / fearful / nervous), and stressed (tense). Regardless of whether QBA is used for assessing the welfare of aquatic or terrestrial species, PC1 appears to typically be influenced heavily by terms that reflect a continuum between the extremes of relaxation and stress / agitation (Sant'Anna and Paranhos da Costa, 2013; Jarvis *et al.*, 2021).

With lights being switched on at precisely 10:30am every morning, this was considered a routine event that could be methodically recorded and expected to help stimulate activity in the fish. This would potentially maximise what expressive characteristics could be captured without causing additional stress to the salmon. The initial 30 seconds were cut out to exclude the salmon's startle responses to the lights, which may have otherwise drowned out any potential differences reflected by the QBA scores. The significant reduction in stocking density, as a result of the sampling, was an additional factor that could influence the QBA scoring in two manners. Firstly, any consistently stark differences in the number of visible salmon between video clips recorded before and after sampling could reveal important differences to the observers that were meant to be blind to treatment. As shown by the snapshots in figure 4-1, however, the relatively small tank size and consistent movement of the salmon helped to make this difference in density far less apparent. Alongside the randomised order of video clips in the scoring sheets, it is unlikely that observers would have been able to pick up on the difference. An additional consideration to the sampling event is that differences captured in the salmon's behaviour may simply be due to the change in stocking density, rather than any stress caused from the sampling itself. Assuming that the reduction in stocking density played a major role in altering the behaviours of the salmon, the dimensions of the QBA that captured a significant difference were still heavily influenced by terms indicating changes in the relaxation and stress / agitation of the salmon. These changes in behaviour, indicative of increased stress in the salmon, ultimately arose as a result of the sampling event (regardless of whether it was from the anaesthetisation, handling out of water, or change in stocking density) and this was successfully captured by QBA.

The LMEM determined that there was significant variation between observers in the mean scores they attributed to the 63 video clips on each PC. This suggests that observers may have been interpreting and using the ranges within the VASs differently, while still agreeing on the direction in which the scores should change from one video to another. Such an occurrence is not uncommon when multiple individuals use the same continuous scales (Bryce and Bratzke, 2015). In most QBA studies, the directionality of scores, as indicated by Kendall's W, is taken as the most important indicator for inter-observer agreement (Clarke, Pluske and Fleming, 2016; Minero *et al.*, 2018; Jarvis *et al.*, 2021). However, crucial to the aims of this study, the observer effect was accounted for by the LMEM when analysing the treatment effect, and thus a significant difference between PC1 scores was found before and after the stressful challenge.

Previous studies have suggested that significant associations between QBA and other welfare measures help support the validity of QBA as a welfare assessment tool (Minero *et al.*, 2018; Muri *et al.*, 2019; Jarvis *et al.*, 2021; Vasdal *et al.*, 2022). However, as noted by (Wemelsfelder, 2007), the purpose of QBA is to examine subtle expressive aspects of an animal's demeanour in ways that would be otherwise difficult to quantify for other measures of behaviour. It is important to be reminded of the multi-faceted nature of welfare (Stien *et* *al.*, 2013; Noble *et al.*, 2018; Weary and Robbins, 2019), and that QBA should be regarded as a complementary addition to an integrated approach involving various welfare indicators. QBA is thus used with the intention of gaining unique insights into an animal's emotional state in a way that is complementary to other indicators, allowing for a more comprehensive evaluation of animal welfare (Wemelsfelder, 2007; Jarvis *et al.*, 2021). Welfare assessments should also aim to minimise redundancies and include measures that are, at least to some degree, independent from each other (Botreau *et al.*, 2007). As there were significant differences in both PC1 scores and feed intake before and after the stressful challenge, and yet they were not correlated with each other, these results should further support the notion of QBA being a unique welfare assessment tool. In somewhat of a contrast to this, darting scores showed a moderately positive correlation to PC1 scores. Put simply, as the salmon were observed to be more unsettled, stressed, spooked / skittish, and agitated, there was a corresponding increase in the frequency and/or intensity of darting events. However, the darting scores alone showed no treatment effect from the stressful challenge. While these two measures were not entirely independent from one another, QBA was capable of capturing a significant treatment effect when the darting scores could not. This finding highlights the sensitivity of QBA, indicating that the PC1 scores were more capable of capturing the effects of the stressful challenge on the salmon's welfare than the darting scores.

PC2 and PC4 showed acceptable inter-observer reliability, explaining proportions of variation that were comparable to other studies applying QBA to terrestrial animals (Temple *et al.*, 2011; Fleming *et al.*, 2015; Minero *et al.*, 2018; Vasdal *et al.*, 2022). For PC2, the only positively loading QBA term was relaxed, with the main negatively loading terms being energetic, purposeful, and inquisitive. This meant that PC2 mainly reflected the salmon's degree of relaxation against 'high energy'; lower PC2 scores reflected more lively, energetic salmon. PC4 was characterised by terms that reflected a shift in how "harmonious" or "consistent" the behaviour of the salmon was as a collective (i.e., cohesive vs. lost / disoriented). PC3 explained one third of the proportion of variation explained by PC1, with poor inter-observer reliability. The terms most heavily loaded for this dimension (indifferent and purposeful) may help partially explain this inconsistency between observers. Such terms could have been more difficult to perceive and assess in salmon, in comparison to the terms used within PC1.

There was no statistically significant difference between the pre- and post- sampling event stages in PC2 or PC4. Sampling was specifically chosen as a presumably intrusive, stressful event, with the intentions of then assessing QBA's ability to detect the putative impacts of such an event on the salmons' emotional state. Considering the terms used to characterise

PC2 and PC4, these dimensions may be relevant for fish welfare under the context of different "treatments", which instead incite reactions that are outside of the typical responses to standard stressors. For example, the "lively, energetic" dimension of PC2 might be suitable for assessing the potential benefits of environmental enrichment, whereas the "harmonious" dimension of PC4 may reflect potential disruptions to the shoaling/schooling behaviours of salmon after transportation/transfer to new enclosures. Considering that the most relevant dimension in the context of this study (PC1) reflects a combined shift in both valence (positive – negative) and energy (low – high), this dimension could be of significant use for on-farm welfare assessments of Atlantic salmon. Additional research is needed to further explore and validate the relevance of other dimensions found in this study (i.e., PC2 and PC4), under different experimental treatments, to expand the potential applications of QBA for salmon welfare assessments.

Integrating QBA into future welfare assessments (for research or farming) will first require appropriate training in the observing, scoring, and understanding of terms involved (Clarke, Pluske and Fleming, 2016; Grosso *et al.*, 2016). While this may require a significant initial investment towards developing the observers' assessment capabilities, doing so will help ensure acceptable inter-observer reliability and, over the long term, help with the integration of a unique and efficient welfare assessment tool (Jarvis *et al.*, 2021). Welfare assessments that include QBA have the advantage of evaluating emotional states of the animals, and the consequent monitoring of positively valenced terms (e.g., content, relaxed, inquisitive, cohesive, purposeful, energetic etc.) also allows for the consideration of positive aspects of fish welfare.

The various ways in which sampling can cause stress and impair fish welfare demonstrates another advantage with implementing QBA; as a non-intrusive method of welfare assessment. QBA avoids any negative impacts from its measurement, an issue that is inherent in many animal-based measures. A large proportion of animal-based measures of welfare are also retrospective, only identifying problems long after they have occurred (Noble *et al.*, 2018). Analyses of behavioural expression could help minimise this delay, perhaps even to the point of providing early warning signs for pre-clinical health issues (Dawkins, 2003). Through virtue of being able to assess behavioural expressions through video monitoring, QBA is also capable of being carried out remotely. Considering the remote locations in which these salmon are often kept (Natural Scotland, 2016), as well as issues surrounding monitoring when site access is limited, this feature provides a significant advantage. The need for such welfare monitoring tools was highlighted to the Scottish salmon farming sector when farm staff were restricted from accessing their sites during the 2020 COVID-19 pandemic, and in-person audits for welfare certification schemes had to be replaced with virtual assessments for two months (FishFarmingExpert, 2020; Murray *et al.*, 2021). During a recent industry survey carried out within the salmon farming sector, various professionals employed in the production process ranked the development of remote, non-intrusive welfare indicators as one of the highest research priorities for farmed salmon welfare (Wiese *et al.*, 2023). The effective implementation of QBA on-site would help meet this demand.

4.6 Conclusion

This is the first study to demonstrate QBA's ability to capture changes in the expressive characteristics of Atlantic salmon following exposure to putatively stressful events. Five observers from various professional backgrounds achieved acceptable inter- and intra-observer reliability in three dimensions of QBA scores. PC1 showed a significant treatment effect, with salmon becoming more unsettled, stressed, spooked / skittish, and agitated after the stressful challenge. Both PC1 scores and feed intake recorded a significant difference before and after the stressful challenge, but were not correlated to each other. PC1 scores showed a moderate positive correlation with darting scores, however the darting scores did not show a significant treatment effect, indicating the QBA scores to be more sensitive to the stressful challenge. These results support QBA's ability to provide unique insights that are relevant to the evaluation of farmed salmon welfare. Future experiments should explore the other dimensions found within QBA (e.g., PC2 and PC4) under different treatment conditions, and across other species of fish, to further investigate QBA's applicability within aquaculture. The results from this study demonstrate that QBA is a promising welfare indicator that, with further research, could act as a time-efficient and complimentary tool for on-farm welfare assessments.

4.7 Acknowledgements

The authors would like to acknowledge the contribution of the multiple staff of Bakkafrost Scotland for their participation in the term generation and Aquascot for participating in the term generation and QBA scoring sessions of this experiment. A special thanks also goes to Maureen Ellis and Pamela Prentice for volunteering to be observers in the QBA scoring sessions. Finally, the authors express their gratitude to Monica Betancor for permitting the filming of the salmon involved in her research at Machrihanish, which greatly facilitated the successful completion of this study.

4.8 References

Ashley, P. J. and Sneddon, L. U. (2008) Pain and fear in fish, in Fish Welfare. *Oxford, UK: Blackwell*, pp. 49–77.

Boissy, A., Manteuffel, G., Jensen, M. B. et al. (2007) Assessment of positive emotions in animals to improve their welfare. Physiology and Behavior, 92, pp. 375–397. doi: 10.1016/j.physbeh.2007.02.003.

Botreau, R., Veissier, I., Butterworth, A. et al. (2007) Definition of criteria for overall assessment of animal welfare. Animal Welfare, 16(2), pp. 225–228.

Des Roches, A. D. B., Lussert, A., Faure, M. et al. (2018) Dairy cows under experimentally-induced Escherichia coli mastitis show negative emotional states assessed through Qualitative Behaviour Assessment. Applied Animal Behaviour Science, 206, pp. 1–11. doi: 10.1016/j.applanim.2018.06.004.

Broom, D. M. (1998) Welfare, stress, and the evolution of feelings. Advances in the Study of Behavior: stress and behaviour, 27, pp. 317–403.

Brown, C. (2015) Fish intelligence, sentience and ethics. Animal Cognition, 18(1), pp. 1–17. doi: 10.1007/s10071-014-0761-0.

Browning, H. (2022) Assessing measures of animal welfare. Biology & Philosophy, 37(36), pp. 1–24.

Browning, H. (2023) Improving welfare assessment in aquaculture. Frontiers in Veterinary Science, pp. 1–10. doi: 10.3389/fvets.2023.1060720.

Bryce, D. and Bratzke, D. (2015) Are introspective reaction times affected by the method of time estimation? A comparison of visual analogue scales and reproduction. Attention, Perception, & Psychophysics, 77, pp. 978–984. doi: 10.3758/s13414-014-0804-2.

Cantalupo, C., Bisazza, A. and Vallortigara, G. (1995) Lateralization of predator-evasion response in a teleost fish (*Girardinus falcatus*). Neuropsychologia, 33(12), pp. 1637–1646.

Chervova, L. S. and Lapshin, D. N. (2011) Behavioral Control of the efficiency of pharmacological anesthesia in fish. Journal of Ichthyology, 51(11), pp. 1126–1132. doi: 10.1134/S0032945211110026.

Clarke, T., Pluske, J. R. and Fleming, P. A. (2016) Are observer ratings influenced by prescription? A comparison of free choice profiling and fixed list methods of qualitative behavioural assessment. Applied Animal Behaviour Science, 177, pp. 77–83. doi: 10.1016/j.applanim.2016.01.022.

Cooper, R. and Wemelsfelder, F. (2020) Qualitative behaviour assessment as an indicator of animal emotional welfare in farm assurance. Livestock, 25(4), pp. 180–183. doi: 10.12968/live.2020.25.4.180.

Darwin, C. (1872) The Expression of the Emotions in Man and Animals, University of Chicago Press: London UK.

Dawkins, M. S. (2003) Behaviour as a tool in the assessment of animal welfare. Zoology, 106(4), pp. 383–387. doi: 10.1078/0944-2006-00122.

Dawkins, M. S. (2004) Using behaviour to assess animal welfare. Animal Welfare, 13(SUPPL.), pp. 3–7.

Dawkins, M. S. (2015) Animal welfare and the paradox of animal consciousness. Advances in the Study of Behavior, 47, pp. 5-38. doi: 10.1016/bs.asb.2014.11.001.

Djordjevic, B., Kristensen, T., Øverli, Ø. et al. (2012) Effect of nutritional status and sampling intensity on recovery after dorsal aorta cannulation in free-swimming Atlantic salmon (*Salmo salar L.*). Fish Physiology and Biochemistry, 38(1), pp. 259–272. doi: 10.1007/s10695-009-9362-2.

Domenici, P. and Blake, R. W. (1997) The kinematics and performance of fish fast-start swimming. Journal of Experimental Biology, 200(8), pp. 1165–1178. doi: 10.1242/jeb.200.8.1165.

Duthie, C. A., Bowen, J. M., Bell, D. J. et al. (2020) Feeding behaviour and activity as early indicators of disease in pre-weaned dairy calves. Animal, 15(3), pp. 1–7. doi: 10.1016/j.animal.2020.100150.

Ellingsen, K., Coleman, G. J., Lund, V. et al. (2014) Using qualitative behaviour assessment to explore the link between stockperson behaviour and dairy calf behaviour. Applied Animal Behaviour Science, 153, pp. 10–17. doi: 10.1016/j.applanim.2014.01.011.

Ellis, T., Turnbull, J. F., Knowles, T. G. et al. (2016) Trends during development of Scottish salmon farming: An example of sustainable intensification?. Aquaculture, 458, pp. 82–99. doi: 10.1016/j.aquaculture.2016.02.012.

FAO (2020) The State of World Fisheries and Aquaculture - Sustainability in action. Rome. doi: https://doi.org/10.4060/ca9229en.

Fife-Cook, I. and Franks, B. (2019) Positive welfare for fishes: Rationale and areas for future Study. Fishes, 4(2), p. 31. doi: 10.3390/fishes4020031.

FishFarmingExpert (2020) RSPCA Assured targets return of farm visits, FishFarmingExpert News. Available at: https://www.fishfarmingexpert.com/article/rspca-assured-takes-first-step-towards-normal-assessments/ (Accessed: 11 October 2021).

Fleming, P. A., Paisley, C. L., Barnes, A. L. et al. (2013) Application of Qualitative Behavioural Assessment to horses during an endurance ride. Applied Animal Behaviour Science, 144(1–2), pp. 80–88. doi: 10.1016/j.applanim.2012.12.001.

Fleming, P. A., Wickham, S. L., Stockman, C. A. et al. (2015) The sensitivity of QBA assessments of sheep behavioural expression to variations in visual or verbal information provided to observers. Animal, 9(5), pp. 878–887. doi: 10.1017/S1751731114003164.

Fleming, P. A., Clarke, T., Wickham, S. et al. (2016) The contribution of qualitative behavioural assessment to appraisal of livestock welfare. Animal Production Science, 56(10), pp. 1569–1578. doi: 10.1071/AN15101.

Franks, B., Ewell, C. and Jacquet, J. (2021) Animal welfare risks of global aquaculture. Science Advances, 7, pp. 1–8.

Fraser, D., Weary, D. M., Pajor, E. A., et al. (1997) A scientific conception of animal welfare that reflects ethical concerns. Animal Welfare, 6(3), pp. 1–19.

Gould, D. (2001) Visual Analog Scale (VAS) - in depth. Journal of Clinical Nursing, (10), pp. 697–706.

Green, T. C. and Mellor, D. J. (2011) Extending ideas about animal welfare assessment to include “quality of life” and related concepts. New Zealand Veterinary Journal, 59(6), pp. 263–271. doi: 10.1080/00480169.2011.610283.

Grosso, L. et al. (2016) On-farm Qualitative Behaviour Assessment of dairy goats in different housing conditions. Applied Animal Behaviour Science, 180, pp. 51–57. doi: 10.1016/j.applanim.2016.04.013.

Hemsworth, P. H. et al. (2015) Scientific assessment of animal welfare. New Zealand Veterinary Journal, 63(1), pp. 24–30. doi: 10.1080/00480169.2014.966167.

Hoffmann, V., Probst, K. and Christinck, A. (2007) Farmers and researchers: How can collaborative advantages be created in participatory research and technology development?. Agriculture and Human Values, 24, pp. 355–368. doi: 10.1007/s10460-007-9072-2.

Huntingford, F. A. et al. (2006) Current issues in fish welfare. Journal of Fish Biology, 68, pp. 332–372. doi: 10.1111/j.1095-8649.2005.01046.x.

IBM Corp. (2021) IBM SPSS Statistics for Windows. Armonk, NY: IBM Corp.

Iversen, M., Finstad, B. and Nilssen, K. J. (1998) Recovery from loading and transport stress in Atlantic salmon (Salmo salar L.) smolts. Aquaculture, 168(1–4), pp. 387–394. doi: 10.1016/S0044-8486(98)00364-0.

Jarvis, S. et al. (2021) Qualitative Behavioral Assessment in juvenile farmed Atlantic salmon (Salmo salar): Potential for on-farm welfare assessment. Frontiers in Veterinary Science, 8, pp. 1–11. doi: 10.3389/fvets.2021.702783.

Jobling, M. et al. (2001) Techniques for measuring feed intake, in Food Intake in Fish. Oxford: Blackwell, pp. 49–87. doi: 10.1002/9780470999516.ch3.

Kaiser, H. F. (1960) The application of electronic computers to factor analysis. Educational and Psychological Measurement, 20, pp. 141–151. Available at: http://dx.doi.org/10.1177/001316446002000116.

Keeling, L. et al. (2013) Welfare Quality® principles and criteria, in Blokhuis, H. et al. (eds) Improving farm animal welfare. Wageningen: Wageningen Academic Publishers, pp. 91–112. doi: https://doi.org/ 10.3920/978-90-8686-770-7_5.

Kristiansen, T. S. et al. (2020) The welfare of fish. Diseases of Aquatic Organisms. Springer. doi: 10.3354/dao075155.

Magurran, A. E. and Pitcher, T. J. (1983) Foraging, timidity and shoal size in minnows and goldfish. Behavioural Ecology and Sociobiology, 12, pp. 147–152. Available at: https://doi.org/10.1007/BF00343206.

Martins, C. I. M. et al. (2012) Behavioural indicators of welfare in farmed fish. Fish Physiology and Biochemistry, 38(1), pp. 17–41. doi: 10.1007/s10695-011-9518-8.

Mason, G. and Mendl, M. (1993) Why is there no simple way of measuring animal welfare?. Animal welfare, 2(4), pp. 301–319.

Mellor, D. J. (2016) Updating animal welfare thinking: moving beyond the “Five Freedoms” towards “A Life Worth Living”. Animals, 6(3), pp. 1–20. doi: 10.3390/ani6030021.

Mendl, M., Burman, O. H. P. and Paul, E. S. (2010) An integrative and functional framework for the study of animal emotion and mood. Proceedings of the Royal Society B: Biological Sciences, 277(1696), pp. 2895–2904. doi: 10.1098/rspb.2010.0303.

Minero, M. et al. (2018) Using qualitative behaviour assessment (QBA) to explore the emotional state of horses and its association with human-animal relationship. Applied Animal Behaviour Science. 204, pp. 53–59. doi: 10.1016/j.applanim.2018.04.008.

Munro, L. A. (2022) Scottish Fish Farm Production Survey 2021. Available at: https://www.gov.scot/publications/scottish-fish-farm-production-survey-2021/pages/5/. Date accessed: 30 June 2023.

Muri, K. et al. (2019) Associations between qualitative behaviour assessments and measures of leg health, fear and mortality in Norwegian broiler chicken flocks. Applied Animal Behaviour Science. 211, pp. 47–53. doi: 10.1016/j.applanim.2018.12.010.

Murray, A. G. et al. (2021) A preliminary assessment of indirect impacts on aquaculture species health and welfare in Scotland during COVID-19 lockdown. Veterinary and Animal Science. 11, pp. 1–5. doi: 10.1016/j.vas.2021.100167.

Napolitano, F., Rosa, G. D. and Grasso, F. (2012) Qualitative behaviour assessment of dairy buffaloes (*Bubalus bubalis*). Applied Animal Behaviour Science, 141, pp. 91–100. doi: 10.1016/j.applanim.2012.08.002.

Natural Scotland (2016) Scotland's aquaculture, Joint Nature Conservation Committee. Available at: http://aquaculture.scotland.gov.uk/map/map.aspx (Accessed: 7 June 2023).

Noble, C., Gismervik, K., Iversen, M. H. et al. (eds.) (2018) Welfare Indicators for farmed Atlantic Salmon: tools for assessing fish welfare http://hdl.handle.net/11250/2575780.

Nomura, M. et al. (2009) Physiology and behaviour of Atlantic salmon (*Salmo salar*) smolts during commercial land and sea transport. Physiology and Behavior. 96(2), pp. 233–243. doi: 10.1016/j.physbeh.2008.10.006.

Oppedal, F., Dempster, T. and Stien, L. H. (2011) Environmental drivers of Atlantic salmon behaviour in sea-cages: A review. Aquaculture. 311(1–4), pp. 1–18. doi: 10.1016/j.aquaculture.2010.11.020.

Pankhurst, N. W. et al. (2008) The relationship between acute stress, food intake, endocrine status and life history stage in juvenile farmed Atlantic salmon, *Salmo salar*, Aquaculture, 275, pp. 311–318. doi: 10.1016/j.aquaculture.2008.01.001.

Rault, J. et al. (2020) The power of a positive human – animal relationship for animal welfare. Frontiers in Veterinary Science, 7, pp. 1–13. doi: 10.3389/fvets.2020.590867.

Rethink Priorities (2019) Rethink Priorities : Submission of Evidence to Animal Welfare (Sentience) Bill. Available at: https://committees.parliament.uk/writtenevidence/37632/pdf/. Date accessed: 15 May 2023.

Rutherford, K. M. D. et al. (2012) Qualitative Behavioural Assessment of emotionality in pigs. Applied Animal Behaviour Science, 139(3–4), pp. 218–224. doi: 10.1016/j.applanim.2012.04.004.

Sandodden, R., Finstad, B. and Iversen, M. (2001) Transport stress in Atlantic salmon (*Salmo salar L.*): Anaesthesia and recovery. Aquaculture Research, 32(2), pp. 87–90. doi: 10.1046/j.1365-2109.2001.00533.x.

Sant'Anna, A. C. and Paranhos da Costa, M. J. R. (2013) Validity and feasibility of qualitative behavior assessment for the evaluation of Nellore cattle temperament. Livestock Science. 157(1), pp. 254–262. doi: 10.1016/j.livsci.2013.08.004.

Santurtun, E., Broom, D. M. and Phillips, C. J. C. (2018) A review of factors affecting the welfare of Atlantic salmon (*Salmo salar*). Animal Welfare, 27(3), pp. 193–204. doi: 10.7120/09627286.27.3.193.

Schreck, C. B., Olla, B. L. and Davis, M. W. (1997) Behavioural responses to stress, in Fish Stress and Health in Aquaculture. Cambridge: Cambridge University Press, p. 155.

Sherrington, C. S. (1906) The Integrative Action of the Nervous System. New York, USA: Scribner.

Sneddon, L. U., Braithwaite, V. A. and Gentle, M. J. (2003) Do fishes have nociceptors? Evidence for the evolution of a vertebrate sensory system. Proceedings of the Royal Society B: Biological Sciences, 270(1520), pp. 1115–1121. doi: 10.1098/rspb.2003.2349.

Stien, L. H. et al. (2013) Salmon Welfare Index Model (SWIM 1.0): A semantic model for overall welfare assessment of caged Atlantic salmon: Review of the selected welfare indicators and model presentation. Reviews in Aquaculture, 5(1), pp. 33–57. doi: 10.1111/j.1753-5131.2012.01083.x.

Temple, D. et al. (2011) Assessment of animal welfare through behavioural parameters in Iberian pigs in intensive and extensive conditions. Applied Animal Behaviour Science, 131(1–2), pp. 29–39. doi: 10.1016/j.applanim.2011.01.013.

Vasdal, G. et al. (2022) Qualitative behaviour assessment as part of a welfare assessment in flocks of laying hens. Applied Animal Behaviour Science, 246. doi: 10.1016/j.applanim.2021.105535.

Veit, W. and Browning, H. (2021) Perspectival pluralism for animal welfare, European Journal for Philosophy of Science. European Journal for Philosophy of Science, 11(1), pp. 1–14. doi: 10.1007/s13194-020-00322-9.

Weary, D. M. and Robbins, J. A. (2019) Understanding the multiple conceptions of animal welfare. Animal Welfare, 28(1), pp. 33–40. doi: 10.7120/09627286.28.1.033.

Welfare Quality® (2009) Welfare Quality® Assessment protocol for poultry (broilers, laying hens). Lelystad, Netherlands.

Wemelsfelder, F. et al. (2001) Assessing the “whole animal”: a free choice profiling approach. Animal Behaviour, 62, pp. 209–220. doi: 10.1006/anbe.2001.1741.

Wemelsfelder, F. (2007) How animals communicate quality of life: The qualitative assessment of behaviour. Animal Welfare, 16, pp. 25–31. doi: 10.1017/S0962728600031699.

Wiese, T. R. et al. (2023) Concerns and research priorities for Scottish farmed salmon welfare – An industry perspective. Aquaculture. 566, p. 739235. doi: 10.1016/j.aquaculture.2023.739235.

Zahl, I. H., Samuelsen, O. and Kiessling, A. (2012) Anaesthesia of farmed fish: Implications for welfare. Fish Physiology and Biochemistry, 38(1), pp. 201–218. doi: 10.1007/s10695-011-9565-1.

CHAPTER 5. General discussion

5.1 Context and aims

Farmed fish welfare is now among the top priorities in a sector continuing to expand to support global food security. The Scottish salmon farming industry is a prime example of this growth, reaching record highs in both overall production and intensification in recent years (Ellis *et al.*, 2016; Munro, 2022). Nurturing farmed salmon welfare plays a crucial role for this industry in meeting ethical responsibilities, fostering public acceptance, and optimising production efficiency and product quality (Broom, 1999; Southgate and Wall, 2001; FSBI, 2002; European Commission, 2016).

Ensuring the proper care of any animal's welfare, however, first requires a detailed understanding of what exactly a good standard of welfare entails. Numerous studies and debates have tried to reach satisfactory conclusions on all matters regarding animal welfare, including: "What is important for welfare?" (Fraser, 2003; Håstein, Scarfe and Lund, 2005; Stien *et al.*, 2013; Santurtun, Broom and Phillips, 2018), "How can it be assessed effectively?" (Fraser, 2009; Noble *et al.*, 2018; Veit and Browning, 2021), and "What constitutes a life worth living?" (Green and Mellor, 2011; Mellor, 2016; Webster, 2016). The challenge with asking such important questions is that there is not always a definitive answer for them. While there is overwhelming evidence that fish are capable of experiencing emotional states that contribute to their welfare, such as pain or fear (Sneddon, Braithwaite and Gentle, 2003a; Chervova and Lapshin, 2011; Kristiansen *et al.*, 2020), the concept of welfare itself is ultimately a philosophical, human construct. The various mental frameworks that have been designed in an attempt to answer the questions above are unavoidably shaped by our own cultural and ethical backgrounds, reflecting personal values and beliefs for what matters most in a "life worth living". Objectively enhancing animal welfare therefore becomes challenging when an inevitable balance must be struck between the various value systems present. This complex, multi-faceted nature of animal welfare is only further amplified when dealing with the anadromous life cycle of Atlantic salmon.

5.1.1 Aims and objectives

The principal aim of this thesis is to provide industry-relevant contributions towards the monitoring and safeguarding of farmed salmon welfare. Recognising the intricacy and human influence involved, this thesis first sets out to gather opinions from those directly involved in salmon husbandry and to evaluate past and current welfare practices within the industry. This provides insights into the following:

1) The relative importance that farm staff place on:
    a. Salmon welfare during different production stages and husbandry practices.
    b. The variety of welfare concerns currently facing farmed salmon.
2) The challenges associated with assessing farmed salmon welfare and providing assurances for what levels of welfare are achieved on-site.
    a. Where research should focus to further develop on-farm welfare assessments.

Obtaining this comprehensive industry overview on salmon welfare then informed the direction of the final experimental chapter. This chapter ultimately validates or further refines a novel on-farm welfare assessment tool that offers the most benefits to the industry. Furthermore, the insights gained provide additional guidance on priorities for future research and on-farm management.

5.2 Conclusions from Chapter 2 and 3

5.2.1 Scottish salmon farming overview – key areas of focus

Participants in Chapter 2 raised a variety of context-dependent welfare concerns, underscoring the need for consistent attention to salmon welfare at every stage of the production process. Nevertheless, key areas of focus were still identified in Chapters 2 and 3. On a broader scale, sea lice, gill health, and water quality were the major areas of concern. Another key priority for salmon welfare, identified in Chapter 2, was ensuring that careful monitoring is in place for husbandry practices where direct contact and disturbance to the salmon is unavoidable. This was particularly the case for general handling, crowding, grading, and interventions, all of which have potential to severely compromise salmon health and welfare (Ashley, 2007; Brown *et al.*, 2008; Powell, Reynolds and Kristensen, 2015). Owing to the risks involved, survey participants also encouraged minimal use of welfare measures that require contact and disturbance to the salmon. This is particularly relevant to salmon already suffering from disease or injury; as they are already under physiological stress, these fish are more susceptible to the cumulative stress that would result from excessive handling prior to any treatment required (Marcos-López *et al.*, 2017).

Chapter 3's investigation into the welfare practices of Scottish salmon farms also provided relevant insights regarding this matter. Overall rates of non-compliance with the RSPCA Assured welfare standards remained relatively constant throughout 2011-2019, with poor record keeping and staff training forming the majority of non-compliances. One category of non-compliance, however, highlighted a growing issue for farm site compliance. This related to farms ensuring that proper care is taken by their staff to minimise hazards posed to salmon welfare during human-animal interactions (including handling and interventions). Echoing the concerns raised in Chapter 2, where contact with or disturbance of the salmon is unavoidable, it is apparent that the refinement or reduction of such practices (wherever possible) would contribute greatly towards improving salmon welfare. As discussed in Chapters 2 and 3, the extent to which human-animal interactions influence salmon welfare is also inherently linked to the quality of staff training (Brown *et al.*, 2008; Størkersen *et al.*, 2021). Outside of improving staff training, the only avenue for mitigating hazards presented by human-animal interactions is through reducing the frequency in which farm staff must be in contact with the salmon.

5.2.2 Overcoming challenges associated with assessing and providing assurances for farmed salmon welfare

As demands for improving salmon welfare grows, so does the need for increased transparency and evidence from the Scottish salmon farming industry (Fidra, 2020; Fidra and Best Fishes, 2022). Legislation aside, additional welfare assurances are exclusively provided through certification schemes including the RSPCA Assured welfare standards (FAWC, 2014; RSPCA, 2021). On-site audits (e.g., those conducted by RSPCA Assured assessors to ultimately evaluate the welfare practices of these farms) thus serve as the sole means for providing evidence regarding what levels of welfare are being achieved for farmed salmon. Preceding the study carried out in Chapter 3, however, there had been limited research into the underlying frameworks for any set of standards providing such assurances. This includes examining:

1) What requirements (or clauses) welfare standards include, and why.
2) How farm sites have complied with the standards, and what insights into their welfare practices this might provide.

Chapter 3 filled this knowledge gap through a comprehensive assessment of the RSPCA Assured welfare standards, and the associated compliance data of certified farm sites from 2011-2019. One of the main findings from this part of the study was that animal-based measures represented less than 2% of the clauses within the welfare standards. As a result, the current form of non-compliance data could only provide insights into what welfare practices farms took to mitigate risks or hazards to salmon welfare. There was insufficient evidence on what actual welfare outcomes were achieved on-site, and no comparisons could be made between different aspects of welfare. For example, comparing estimates of their overall states of physical well-being vs. their mental well-being, or between different production stages (e.g., general levels of welfare achieved at the seawater rearing stage vs.

hatchery stage). Chapter 3 thus demonstrated the important role that direct measures of the salmon's physical of behavioural attributes play in providing evidence on what transpired for their welfare. Despite the RSPCA Assured (and other industry stakeholders) explicitly stating the importance of animal-based measures (Berrill *et al.*, 2012; RSPCA, 2021), the formal inclusion of such measures as auditable clauses (or 'welfare outcome assessments') has so far remained minimal. As a result, this thesis attempted to determine why this was the case.

The issues of practicality and effectiveness when carrying out animal-based measures on-site, highlighted by industry participants in Chapter 2, provided some potential explanations. The overall sentiment regarding these measures, particularly those which require intrusive handling of the salmon (e.g., assessments of abnormalities or physiological measures of stress), was that they posed major risks to their welfare while scoring relatively poorly in terms of on-site practicality and effectiveness. The use of animal-based measures, however, was not regarded as costly or time-consuming if the measures involved were non-intrusive to the salmon. In accordance with minimising human-animal interactions with the salmon, industry participants highlighted the need for further developing operational welfare indicators ('OWIs') that possess the following capabilities:

1) Non-intrusive; their use results in no disturbance of the salmon.
2) Animal-based; directly measures a physical or behavioural attribute of the salmon.
3) Can be carried out remotely; farm staff are not required on-site for its use.

Chapter 3 reached a similar conclusion, attributing the lack of animal-based measures as auditable clauses to issues of practicality and disruptions that would likely arise (to both salmon and farming routines) during the auditing process.

With the potential to address all these issues, behavioural assessments were identified in Chapter 2 as the most promising area for further developing. The potential advantages of incorporating behavioural welfare indicators into on-farm monitoring and management strategies are already well acknowledged (Dawkins, 2003; Huntingford *et al.*, 2006; Oppedal, Dempster and Stien, 2011; Martins *et al.*, 2012; Miller *et al.*, 2020; Barreto *et al.*, 2021; O'Donncha *et al.*, 2021). Behaviours can be monitored and assessed through direct human observation, manual coding of video recordings, or through more automated methods including acoustic monitoring or computer vision techniques (Wemelsfelder and Lawrence, 2001; Martins *et al.*, 2012; Rushen and De Passille, 2012; Terayama *et al.*, 2019; Barreto *et al.*, 2021). Since behaviours can be monitored through video recordings, such assessments can be carried out remotely. Direct measures of animal welfare often come with the cost of either being time-consuming, technically complex, or disturbing the salmon. Behavioural indicators, however, are one of the few that are comparatively fast, non-intrusive, and easy to observe (Dawkins, 2004; Huntingford *et al.*, 2006; Martins *et al.*, 2012). Monitoring changes in behaviour may also provide early warning signs for issues that arise on site (Huntingford *et al.*, 2006; Oppedal, Dempster and Stien, 2011; Wiese *et al.*, 2023). Improving the ability for farm staff to recognise and resolve problems before they severely impact the stock benefits both the salmon and farm productivity. This advantage was also identified by participants in Chapter 2, who commented on the potential benefits (for both salmon welfare and farm production) for further implementing behavioural assessments on-site. Furthermore, behavioural assessments are currently the only tool that allow for any relevant insights into the subjective experiences of animals (Turnbull and Kadri, 2007; Folkedal *et al.*, 2012; Martins *et al.*, 2012; Mellor, 2012; Dawkins, 2015; Zhao, Bao, Zhang, Zhu, Liu, Lu, *et al.*, 2018; Hassan *et al.*, 2019). Behaviours exhibited by an animal are regarded by some as the final product of all its own decision-making processes (Dawkins, 2004; Mendl, Burman and Paul, 2010), and as such are ultimately an "expression of the emotions" (Darwin, 1872).

There are therefore clear advantages associated with behavioural welfare indicators, and yet their formal inclusion within certain welfare assessments (i.e., RSPCA Assured audits) is still minimal (RSPCA, 2021). Various limitations currently facing this class of welfare indicators, listed below, may provide some explanation for this:

5.2.2.1 Logistical issues of monitoring behaviour in aquaculture

Monitoring behaviour in aquaculture systems has typically favoured group-level observations over those of each individual (Millman, 2007; Prunet *et al.*, 2012; Føre *et al.*, 2017; Barreto *et al.*, 2021). This is primarily a result of logistical issues in monitoring such large numbers of fish within a 3-dimensional environment, where complex swimming behaviours and visual obstruction of individuals are frequent (Ye *et al.*, 2016). The issue with relying solely on group-level observations, however, is that they do not always accurately reflect the experiences of individuals, nor can they account for the range of individual-variation present within each group (Martins *et al.*, 2012; Cleasby, Nakagawa and Schielzeth, 2015; Barreto *et al.*, 2021; Daigle and Siegford, 2023). A number of methods for analysing fish behaviour also rely on a light source, and the use of unnatural illumination to ensure sufficient quality in videos may also influence fish behaviour (Bruning, Holker and Wolter, 2011).

5.2.2.2 Quantifying behavioural indicators and time constraints

When applied to welfare assessments, many behavioural indicators are difficult to quantify without appropriate training and are dependent on the motivation and skills of the observer

(Noble *et al.*, 2018). In addition, obtaining actionable insights from an animal's behaviour is often only achievable after further analysis of, e.g., collected video data (Noble *et al.*, 2018), and thus not time-efficient for formal welfare assessments. This is particularly relevant for audits conducted by certification schemes, where assessors may have hundreds of clauses to evaluate in a single day.

5.2.2.3 Potential misinterpretation of behaviours for welfare assessments

When evaluating changes in distinct, physical behaviours (e.g., swimming speeds), there are also risks of misinterpreting how this relates to the animal's welfare. Specific behavioural responses are often considered as either normal coping activities, or abnormal / maladaptive responses; however, the differences between the two are frequently unclear (Martins *et al.*, 2012). Even in situations where the same distinct behavioural response is exhibited, context can be crucial (Ruiz-gomez *et al.*, 2008). For example, elevated swimming speeds during feeding can indicate underfeeding for various aquaculture species (Huse and Skiftesvik, 1985; Björnsson, 1993; Andrew *et al.*, 2004). Conversely, the same response may simply indicate an increased motivation to feed, and be an aspect of the fish's foraging strategy (Kristiansen and Ferno, 2007). The potential dichotomy in how single behaviours can be interpreted as either poor or good welfare demonstrates the importance of having not only species-specific, but context-specific behavioural welfare indicators (Herbert and Steffensen, 2005; Martins *et al.*, 2012).

A key requirement in achieving the main aim of this thesis was the validation of a novel on-farm welfare assessment tool. Findings from Chapters 2 and 3 emphasized the value of further developing non-intrusive, animal-based welfare indicators that can be carried out remotely. By validating a behavioural welfare indicator with these capabilities, this thesis would take a significant step towards the practicality and effectiveness of on-farm welfare assessments. However, there are clear limitations (mostly practical) that currently hinder the full potential for behavioural indicators to fulfil this role. It was therefore vital that this thesis not only validated a welfare assessment tool that aligns with the key findings from Chapters 2 and 3, but also addressed any of the associated limitations and propose potential solutions for them.

5.3 Chapter 4 - QBA's potential for farmed salmon welfare assessments

Through observing the "whole-animal's" behavioural expressions (including its appearance and interactions with others and the surrounding environment), QBA serves as an integrative welfare indicator that reveals insights into the animal's emotional state (Wemelsfelder, 2007;

Ellingsen *et al.*, 2014; Rose and Riley, 2019; Cooper and Wemelsfelder, 2020; Vasdal *et al.*, 2022). Through virtue of assessing behavioural expressions, QBA benefits from practicality and effectiveness for on-farm welfare assessments (e.g., non-intrusive, animal-based, and can be carried out remotely). QBA has also been used to successfully detect early clinical signs of disease, e.g., mastitis in dairy cows (De Boyer des Roches *et al.*, 2018), and could therefore play an important role in early warning systems for farmed salmon. QBA's application also provides a number of unique value propositions. The expressive characteristics used within QBA are intended to cover the full range of both negative and positive emotions (Jarvis et al., 2021; Browning, 2023), thereby allowing QBA to capture positive aspects of welfare (Rose and Riley, 2019). There is therefore the potential to quantify the likelihood that salmon have positive welfare experiences under different environments / husbandry practices. QBA may then also allow for an assessment of the efficacy of enrichment strategies (Rose and Riley, 2019). QBA's approach is also useful for achieving large-scale, long-term datasets regarding changes in behavioural patterns, providing insights into the appropriateness of other husbandry and management regimes (Rose and Riley, 2019).

Despite the advantages of using QBA as a welfare indicator, there has been limited research examining its application within aquaculture (Jarvis *et al.*, 2021). Until Chapter 4, no study had yet examined QBA's ability to capture changes in the behavioural expression of Atlantic salmon (or any other species of fish) following exposure to a putatively stressful challenge. Furthermore, no study had compared QBA scores with other welfare indicators in this context. Chapter 4 reports on the first study to demonstrate QBA's abilities, capturing a significant treatment effect when darting scores did not. While feed intake was also able to capture a significant treatment effect, the two indicators were not correlated with each other. This further supported the notion that QBA provides unique insights into salmon welfare. The findings from Chapter 4 represent a direction taken towards validating a welfare assessment tool that is novel to aquaculture, based on a comprehensive industry overview obtained in Chapters 2 and 3. This application of QBA demonstrates its potential as either an auditable clause within on-farm welfare assessments, or as an OWI for routine monitoring. The full realisation of this potential, however, is contingent upon addressing the limitations that behavioural welfare indicators currently face. Fortunately, behavioural assessments are well positioned to leverage a suite of emerging technologies to overcome these limitations.

5.4 Automation within behavioural assessments

The inability to collect actionable data on-site, in a time-efficient manner, directly limits how farm management strategies can resolve issues as they arise (Bell *et al.*, 2022). To aid with this, the salmon farming industry has begun adopting a ‘precision farming’ approach, applying a suite of technologies to facilitate automated, real-time monitoring and analysis of fish behaviour, welfare, environmental impacts, and production parameters (Berckmans, 2017; Føre *et al.*, 2017; Erp-van der Kooij and Rutter, 2020; O’Donncha *et al.*, 2021). Utilising data-driven insights within salmon welfare will help increase the capabilities of OWIs that are already developed (Barreto *et al.*, 2021; O’Donncha *et al.*, 2021). Behavioural welfare indicators are particularly well positioned to take advantage of these innovations (Valletta *et al.*, 2017; Christin, Hervet and Lecomte, 2019; O’Donncha *et al.*, 2021). These innovations not only address previously discussed limitations, but further reinforce the advantages of behavioural assessment. Underwater video monitoring systems, either fixed of mounted on underwater vehicles, are becomingly increasingly common for monitoring salmon behaviour and welfare (Shortis *et al.*, 2016; Bjerkeng *et al.*, 2021; Bell *et al.*, 2022). Video monitoring also allows for footage to be reviewed remotely by farm staff, or potentially accessible for auditors if the certification schemes and farms involved agree to this (enabling such assessments to be carried out remotely). Through virtue of using video recordings, such footage can be reviewed remotely by farm staff or potentially accessed by auditors if the certification scheme and farms involved agree to this. These monitoring systems, however, face a number of challenges including visual obstructions, tracking complex individual behaviours amongst large groups, poor light conditions, time-constraints for video analysis, training requirements for behavioural assessments, and the potential for observer bias (Pinkiewicz, Purser and Williams, 2011; Barnard *et al.*, 2016; Saberioon and Cisar, 2016; Saberioon *et al.*, 2017; Noble *et al.*, 2018).

Computer vision and machine learning have found real-world applications in facilitating non-intrusive, automatic methods for on-site monitoring of fish behaviour (Kane, Salierno and Gipson, 2008; Kohda *et al.*, 2015; Saberioon *et al.*, 2017; Wang and Takeuchi, 2017; Terayama *et al.*, 2019). Computer vision (i.e., machine vision systems) can be defined as the construction of explicit information and meaningful descriptions of physical objects via image analysis (Glinski, Horabik and Lipiec, 2011). Innovations in this area of computer science have grown rapidly in recent years, becoming more sensitive, powerful and cheaper alongside developments in digital cameras and speeds of computer-based processing (Zion, 2012; Saberioon *et al.*, 2017). Machine learning, in general, refers to a variety of algorithms that can automatically generate predictive models by detecting patterns of data (Christin, Hervet and Lecomte, 2019), and its relevance will be further explained in a later section.

5.4.1 Automated detection and tracking of behaviours

Video tracking involves the tracking of moving objects and monitoring their activities through processing the sequence of images captured in a video recording (Maggio and Cavallaro, 2011). The application of this technology to automatically quantify behavioural parameters has been made possible through a number of methods involving image and motion analysis techniques (Patullo, Jolley-Rogers and Macmillan, 2007; Grubich, Rice and Westneat, 2008; Duarte, Reig and Oca, 2009). An advantage of automated systems that detect behavioural changes via computer imaging is its efficiency after implementation; once installed, no labour is required to obtain behavioural information and action can be taken when the system detects abnormal deviations from normal levels of fish activity (Xia *et al.*, 2016; Barreto *et al.*, 2021). Examples of this technology have already been used to systematically detect subtle behavioural changes (indicative of stress within a group) of Nile tilapia (Zhao, Bao, Zhang, Zhu, Liu and Lu, 2018). Sonar and optic video imaging have also been used, in combination with a deep neural network, to facilitate regular observations of fish groups under sub-optimal lighting conditions and potentially detect behavioural parameters (Terayama *et al.*, 2019). Another low-light, relatively inexpensive option is near-infrared (NIR) imaging, which uses the electromagnetic spectrum between visible and middle infrared light (Lin *et al.*, 2018; Wang, 2019; Barreto *et al.*, 2021).

The majority of studies mentioned above have tested their tracking systems under laboratory conditions, and it is likely that real-world conditions will present additional challenges. Tidal, a subsidiary of Google's parent group Alphabet, have developed a novel underwater monitoring system which incorporates a set of computer vision tools to capture farmed salmon behaviour (Gairn, 2023; Tidal -X, 2023). Tidal has tested the capabilities of this monitoring system within offshore farms in Norway, claiming it is now capable of continuously detecting and tracking individual behaviours under rough oceanic conditions, as well as modelling behaviours over time to provide new insights into farmed fish welfare (Tidal -X, 2023).

5.4.2 Automated analysis of behaviour

The innovations described thus far have largely referred to the automatic detection and tracking of behaviours, including the potential to detect changes in these behaviours. However, in order to enable in-depth analyses of these behaviours (to the point where actionable data on salmon welfare can be obtained automatically), further advancements are required. When combined with computer vision systems, machine learning tools have specific relevance to ecology and behavioural sciences through their capabilities of analysing the complex, nonlinear data encountered in this field (Olden, Lawler and Poff, 2008). Machine learning can occur under ‘supervised learning’, where labelled datasets (e.g., videos showing a variety of the target object’s behaviour, in this case salmon) are first given to the artificial agent so that they can train themselves to associate the labels with the examples provided (Christin, Hervet and Lecomte, 2019). Following this ‘training’, these artificial agents can then recognise and identify these objects’ (salmon) behaviours in completely new datasets (Lecun, Bengio and Hinton, 2015). However, for the purpose of performing tasks like behavioural assessment, providing only labels within conventional machine learning is insufficient: the user must also specify within the computer’s algorithm precisely what to look for (Christin, Hervet and Lecomte, 2019). For example, in order to identify the salmon and their behaviours in any sequence of images, the algorithm requires specific properties (e.g., a salmon’s shape, colour, size, patterning and finally, specific behaviours) to be explicitly stated to it down to the patterns of pixels (Christin, Hervet and Lecomte, 2019). In contrast, deep learning methods bypass this step; these algorithms are able to automatically detect and extract the required features or properties from the data (i.e., video recordings) provided (Christin, Hervet and Lecomte, 2019). This means that users only need to tell a deep learning algorithm that salmon are present in the footage and, given enough examples, these systems can potentially determine what a salmon (or what various salmon performing certain behaviours) looks like. Convolutional neural networks (CNN) and Deep Reinforcement Learning (DRL) are powerful machine learning technologies that have successfully been applied to various computer vision applications including object identification and tracking, video analysis, and behavioural recognition and classification (Kabra *et al.*, 2013; Qin, Yu and Zhao, 2018; Christin, Hervet and Lecomte, 2019; Le *et al.*, 2021). Deep learning could eventually play a significant role in on-farm welfare assessments by providing fast, objective, practical, and reliable ways to analyse enormous amounts of monitoring data (Christin, Hervet and Lecomte, 2019). An added benefit of automated analyses is that they are not influenced by observer bias, which is particularly relevant for behavioural assessments (Martinez-de Dios, Serna and Ollero, 2003; Polonschii, Bratu and Gheorghiu, 2013; Saberioon *et al.*, 2017).

Incorporating deep learning solutions is no minor task, and the initial investments required for obtaining the relevant training datasets (i.e., thousands of hours of footage), the time taken in training the artificial agents, the development complexity and computing power are all aspects that must be considered before undertaking the deep machine learning approach (Christin, Hervet and Lecomte, 2019). However, a similar approach has already been successfully implemented with human behaviours (McFarland, 2022). Before any of the aforementioned technologies could be successfully implemented within the Scottish salmon farming sector, their applications must first be strictly validated and their use should ultimately minimise the production costs, time requirements, and intrusiveness of on-farm welfare assessments (Barreto *et al.*, 2021). Future research should focus on commercial scale testing, alongside industry consultation, to determine exactly how feasible and beneficial these innovations could be for on-site use (Barreto *et al.*, 2021).

5.4.3 Near-term opportunities and outlook for the future

In the meantime, more short-term steps can be taken to facilitate behavioural welfare indicators. For example, with QBA; in the same way that Visual Analogue Scales (VASs) were created digitally and used in SurveyMonkey to automatically quantify QBA scores in Chapter 4, smart phone applications are already capable of allowing farm staff and auditors to immediately quantify their observations of the salmon's behavioural expressions (Ravenscraft, 2022). Pending further developments in automated data analytics platforms, there is also potential for statistical analyses to be carried out with minimal labour and time costs to farm staff. Data wrangling (i.e., the preprocessing of raw data into a structured and usable format) is often an essential first step in statistical analysis, but requires considerable time for understanding, cleaning, and preparing the data in order to identify any meaningful patterns and insights (Williams *et al.*, 2022). 'Semi-automated' tools for real-world data wrangling have already been developed, making the process of data analytics significantly less time-consuming and laborious (Williams, 2022).

With further developments in machine learning and computer vision technologies, there is potential for the majority of behavioural assessments (perhaps even including QBA) to not only function as a practical OWI for welfare monitoring and audits, but to also act as early-warning systems that require minimal time and effort from farm staff (Rushen and De Passille, 2012). Automating the QBA process specifically, through such algorithms, would inevitably require breaking down the expressive characteristics of salmon into quantifiable features / patterns. To some degree, this would lead to QBA operating more as a reductionist tool, deviating from its original purpose as a holistic assessment. However, it is worth noting that QBA performed through human observation is also not entirely free of reductionism itself: humans inherently categorise and label specific expressions based on their own interpretations and preconceptions. The distinction between these two approaches will partly depend on what differences there are in the cognitive and perceptual capabilities of humans versus machines, which in the near future may not be as substantial as commonly assumed (Korteling *et al.*, 2021). As aquaculture progresses further into the realm of big data, the industry's reliance on artificial intelligence to analyse data will become more and more prevalent (Christin, Hervet and Lecomte, 2019). There will then be the task of recruiting individuals with the appropriate programming and mathematical skills and tools, which will likely require increased collaboration across disciplines (Carey *et al.*, 2019). A more in-depth, connected network of computer scientists within aquaculture (both through academia and the commercial sector) could also lead to new synergies and approaches to data classification and analyses, providing new insights for fundamental and applied research in fish health and welfare (Christin, Hervet and Lecomte, 2019). To meet the anticipated need for enhanced collaboration, increased sharing of datasets, codes, and research findings will be crucial for making substantial progress in this field (Christin, Hervet and Lecomte, 2019).

The results from this study outline a promising path through which behavioural welfare indicators can be developed further to advance the practicality and effectiveness of on-farm welfare assessments. In particular, this study has demonstrated QBA's ability to provide unique, reliable insights into salmon welfare while capturing the impacts from a putatively stressful event. Various avenues are available, both in the near and long term, for improving behavioural assessments like QBA to the point where they can provide actionable information of farmed salmon welfare in a practical and time-efficient manner.

5.5 References

Andrew, J. E. et al. (2004) The effect of competition on the feeding efficiency and feed handling behaviour in gilthead sea bream (*Sparus aurata L.*) held in tanks. Aquaculture, 232, pp. 317–331. doi: 10.1016/S0044-8486(03)00528-3.

Ashley, P. J. (2007) Fish welfare: Current issues in aquaculture. Applied Animal Behaviour Science, 104(3–4), pp. 199–235. doi: 10.1016/j.applanim.2006.09.001.

Barnard, S. et al. (2016) Quick, Accurate, Smart: 3D Computer Vision Technology Helps Assessing Confined Animals' Behaviour. PLoS ONE, pp. 1–20. doi: 10.1371/journal.pone.0158748.

Barreto, M. O. et al. (2021) Emerging indicators of fish welfare in aquaculture. Reviews in Aquaculture, pp. 1–19. doi: 10.1111/raq.12601.

Bell, J. L. et al. (2022) Environmental monitoring tools and strategies in salmon net-pen aquaculture, Integrated Environmental Assessment and Management, 18(4), pp. 950–963. doi: 10.1002/ieam.4622.

Berckmans, D. (2017) General introduction to precision livestock farming. Animal Frontiers, 7(1), pp. 6–11. doi: 10.2527/af.2017.0102.

Berrill, I. K. et al. (2012) Achieving consensus on current and future priorities for farmed fish welfare: A case study from the UK. Fish Physiology and Biochemistry, 38(1), pp. 219–229. doi: 10.1007/s10695-010-9399-2.

Bjerkeng, M. et al. (2021) ROV navigation in a fish cage with laser-camera triangulation. Journal of Marine Science and Engineering, 9(79), pp. 1–16.

Björnsson, B. (1993) Swimming speed and swimming metabolism of Atlantic cod (Gadus morhua) in relation to available food: a laboratory study. Canadian Journal of Fisheries and Aquatic Sciences, 50(12), pp. 2542–2551.

De Boyer des Roches, A. et al. (2018) Dairy cows under experimentally-induced *Escherichia coli* mastitis show negative emotional states assessed through Qualitative Behaviour Assessment. Applied Animal Behaviour Science. 206, pp. 1–11. doi: 10.1016/j.applanim.2018.06.004.

Broom, D. M. (1999) Fish Welfare and the public perception of farmed fish. Proceedings of Aquavision, 98, pp. 89-91.

Brown, D. M. et al. (2008) Animal welfare aspects of husbandry systems for farmed Atlantic salmon - Scientific opinion of the panel on animal health and welfare. The EFSA Journal, 6(7). doi: 10.2903/j.efsa.2008.796.

Browning, H. (2023) Improving welfare assessment in aquaculture. Frontiers in Veterinary Science, 10, pp. 1–10. doi: 10.3389/fvets.2023.1060720.

Bruning, A., Holker, F. and Wolter, C. (2011) Artificial light at night: implications for early life stages development in four temperate freshwater fish species. Aquatic Sciences, 73, pp. 143–152. doi: 10.1007/s00027-010-0167-2.

Carey, C. C. et al. (2019) Enhancing collaboration between ecologists and computer scientists: lessons learned and recommendations forward. Ecosphere, 10(5). doi: 10.1002/ecs2.2753.

Chervova, L. S. and Lapshin, D. N. (2011) Behavioral control of the efficiency of pharmacological anesthesia in Fish. Journal of Ichthyology, 51(11), pp. 1126–1132. doi: 10.1134/S0032945211110026.

Christin, S., Hervet, E. and Lecomte, N. (2019) Applications for deep learning in ecology. Methods in Ecology and Evolution, 10, pp. 1632–1644. doi: 10.1111/2041-210X.13256.

Cleasby, I. R., Nakagawa, S. and Schielzeth, H. (2015) Quantifying the predictability of behaviour: statistical approaches for the study of between-individual variation in the within-individual variance. Methods in Ecology and Evolution, 6, pp. 27–37. doi: 10.1111/2041-210X.12281.

Cooper, R. and Wemelsfelder, F. (2020) Qualitative behaviour assessment as an indicator of animal emotional welfare in farm assurance. Livestock, 25(4), pp. 180–183. doi: 10.12968/live.2020.25.4.180.

Daigle, C. and Siegford, J. (2023) Welfare Quality® parameters do not always reflect hen behaviour across the lay cycle in non-cage laying hens. Animal Welfare, 23(4), pp. 423–434. doi: doi:10.7120/09627286.23.4.423.

Darwin, C. (1872) The Expression of the Emotions in Man and Animals, University of Chicago Press: London UK.

Dawkins, M. S. (2003) Behaviour as a tool in the assessment of animal welfare. Zoology, 106(4), pp. 383–387. doi: 10.1078/0944-2006-00122.

Dawkins, M. S. (2004) Using behaviour to assess animal welfare. Animal Welfare, 13(SUPPL.), pp. 3–7.

Dawkins, M. S. (2015) Animal welfare and the paradox of animal consciousness. Advances in the Study of Behavior, 47, pp. 5-38. doi: 10.1016/bs.asb.2014.11.001.

Duarte, S., Reig, L. and Oca, J. (2009) Aquacultural engineering measurement of sole activity by digital image analysis. Aquacultural Engineering, 41, pp. 22–27. doi: 10.1016/j.aquaeng.2009.06.001.

Ellingsen, K. et al. (2014) Using qualitative behaviour assessment to explore the link between stockperson behaviour and dairy calf behaviour. Applied Animal Behaviour Science, 153, pp. 10–17. doi: 10.1016/j.applanim.2014.01.011.

Ellis, T. et al. (2016) Trends during development of Scottish salmon farming: An example of sustainable intensification?. Aquaculture, 458, pp. 82–99. doi: 10.1016/j.aquaculture.2016.02.012.

Erp-van der Kooij, E. V. and Rutter, S. M. (2020) Using precision farming to improve animal welfare, CAB Reviews: Perspectives in Agriculture. Veterinary Science, Nutrition and Natural Resources, 15(51). doi: 10.1079/PAVSNNR202015051.

European Commission (2016) Attitudes of Europeans towards Animal Welfare - Special Eurobarometer. Available at: https://europa.eu/eurobarometer/surveys/detail/2096 (Accessed 10 January 2023).

FAWC (2014) Opinion on the Welfare of Farmed Fish. *FAWC report,* pp. 1-40. Available at: https://assets.publishing.service.gov.uk/media/5a7eb32340f0b62305b829c4/Opinion_on_the_welfare_of_farmed_fish_at_the_time_of_killing.pdf (Accessed: 11 October 2021).

Fidra (2020) Fidra survey reveals: UK consumers will pay more for 'sustainably produced' Scottish salmon & feel ill-informed, Best Fishes. Available at: https://www.bestfishes.org.uk/fidra-survey-reveals-uk-consumers-will-pay-more-if-scottish-salmon-is-sustainably-produced-but-dont-feel-well-informed/ (Accessed: 3 June 2023).

Fidra and Best Fishes (2022) Scottish Salmon Farming - Consumer survey results. Available at: https://www.bestfishes.org.uk/wp-content/uploads/Consumer-survey-2021-Report_FINAL.pdf.

Folkedal, O. et al. (2012) Food anticipatory behaviour as an indicator of stress response and recovery in Atlantic salmon post-smolt after exposure to acute temperature fluctuation. Physiology and Behavior,105(2), pp. 350–356. doi: 10.1016/j.physbeh.2011.08.008.

Føre, M. et al. (2017) Precision fish farming: A new framework to improve production in aquaculture. Biosystems Engineering, 173, pp. 176–193. doi: 10.1016/j.biosystemseng.2017.10.014.

Fraser, D. (2003) Assessing animal welfare at the farm and group level: The interplay of science and values. Animal Welfare, 12(4), pp. 433–443.

Fraser, D. (2009) Assessing animal welfare: Different philosophies, different scientific approaches. Zoo Biology, 28(6), pp. 507–518. doi: 10.1002/zoo.20253.

FSBI (2002) Fish Welfare, Briefing Paper 2, Fisheries Society of the British Isles. Fish & Shellfish Immunology. Available at https://fsbi.org.uk/wp-content/uploads/2018/02/brief-welfare-refs.pdf (Accessed: 15 June 2023).

Gairn, L. (2023) Google group's Tidal platform opened to global aquaculture market, We Are Aquaculture. Available at: https://weareaquaculture.com/news/google-tidal-platform-global-aquaculture-market/36815/ (Accessed: 20 June 2023).

Glinski, J., Horabik, J. and Lipiec, J. (2011) Encyclopedia of Agrophysics. Institute of Agrophysics. Available at: https://www.researchgate.net/profile/Jianhui-Zhang-7/publication/301992634_Tillage_Erosion/links/5817e33a08ae90acb2429e47/Tillage-Erosion.pdf.

Green, T. C. and Mellor, D. J. (2011) Extending ideas about animal welfare assessment to include “quality of life” and related concepts. New Zealand Veterinary Journal, 59(6), pp. 263–271. doi: 10.1080/00480169.2011.610283.

Grubich, J. R., Rice, A. N. and Westneat, M. W. (2008) Functional morphology of bite mechanics in the great barracuda (*Sphyraena barracuda*). Zoology, 111, pp. 16–29. doi: 10.1016/j.zool.2007.05.003.

Hassan, W. et al. (2019) Internet of Fish: Integration of acoustic telemetry with LPWAN for efficient real-time monitoring of fish in marine farms. Computers and Electronics in Agriculture, 163, pp. 1–9. doi: 10.1016/j.compag.2019.06.005.

Håstein, T., Scarfe, A. D. and Lund, V. L. (2005) Science-based assessment of welfare: Aquatic animals. OIE Revue Scientifique et Technique, 24(2), pp. 529–547. doi: 10.20506/rst.24.2.1590.

Herbert, N. A. and Steffensen, J. F. (2005) The response of Atlantic cod, *Gadus morhua*, to progressive hypoxia: fish swimming speed and physiological stress. Marine Biology, 147, pp. 1403–1412. doi: 10.1007/s00227-005-0003-8.

Huntingford, F. A. et al. (2006) Current issues in fish welfare. Journal of Fish Biology, 68, pp. 332–372. doi: 10.1111/j.1095-8649.2005.01046.x.

Huse, I. and Skiftesvik, A. B. (1985) Qualitative and quantitative behaviour studies in starving and feeding turbot (*Scopthalmus maximus L*.) larvae, International Council for the Exploration of the Sea, 38, pp. 1-14.

Jarvis, S. et al. (2021) Qualitative Behavioral Assessment in Juvenile Farmed Atlantic Salmon (*Salmo salar*): Potential for On-Farm Welfare Assessment. Frontiers in Veterinary Science, 8, pp. 1–11. doi: 10.3389/fvets.2021.702783.

Kabra, M. et al. (2013) JAABA: interactive machine learning for automatic annotation of animal behavior. Nature Methods, 10, pp. 64–67. doi: https://doi.org/10.1038/nmeth.2281.

Kane, J. D., Salierno, A. S. and Gipson, G. T. (2008) Quantitative movement analysis of social behavior in mummichog, *Fundulus heteroclitus*. Journal of Ethology, 26, pp. 35–42. doi: 10.1007/s10164-006-0027-7.

Kohda, M. et al. (2015) Facial Recognition in a Group-Living Cichlid, PLoS ONE, 10(11), pp. 1–12. doi: 10.1371/journal.pone.0142552.

Korteling, J. E. et al. (2021) Human-versus artificial intelligence. Frontiers in Artificial Intelligence, 4, pp. 1–13. doi: 10.3389/frai.2021.622364.

Kristiansen, T. S. et al. (2020) The welfare of fish. Animal Welfare, 75(2), pp. 155-158. doi: 10.3354/dao075155.

Kristiansen, T. S. and Ferno, A. (2007) Individual behaviour and growth of halibut (*Hippoglossus hippoglossus L*.) fed sinking and floating feed: Evidence of different coping styles. Applied Animal Behaviour Science, 104, pp. 236–250. doi: 10.1016/j.applanim.2006.09.007.

Le, N. et al. (2021) Deep Reinforcement Learning in Computer Vision: A Comprehensive Survey, ArXiv - Computer Science. Artificial Intelligence Review, pp. 1–103.

Lecun, Y., Bengio, Y. and Hinton, G. (2015) Deep learning. Nature, 521, pp. 436–444. doi: 10.1038/nature14539.

Lin, K. et al. (2018) Three-dimensional location of target fish by monocular infrared imaging sensor based on a L–z correlation model. Infrared Physics and Technology, 88, pp. 106–113. doi: 10.1016/j.infrared.2017.11.002.

Maggio, E. and Cavallaro, A. (2011) Video Tracking: Theory and Practice. Chichester: John Wiley & Sons Ltd.

Marcos-López, M. et al. (2017) Oxidative stress is associated with late-stage amoebic gill disease in farmed Atlantic salmon (*Salmo salar L.*). Journal of Fish Diseases, 41, pp. 1–6. doi: 10.1111/jfd.12699.

Martinez-de Dios, J. R., Serna, C. and Ollero, A. (2003) Computer vision and robotics techniques in fish farms. Robotica, 21(3), pp. 233–243. doi: doi:10.1017/S0263574702004733.

Martins, C. I. M. et al. (2012) Behavioural indicators of welfare in farmed fish. Fish Physiology and Biochemistry, 38(1), pp. 17–41. doi: 10.1007/s10695-011-9518-8.

McFarland, A. (2022) Computer vision technique predicts human behaviour from videos, Artificial Intelligence. Available at: https://www.unite.ai/computer-vision-technique-predicts-human-behavior-from-videos/. Date accessed: 30 June 2023.

Mellor, D. J. (2012) Animal emotions, behaviour and the promotion of positive welfare states. New Zealand Veterinary Journal, 60(1), pp. 1–8. doi: 10.1080/00480169.2011.619047.

Mellor, D. J. (2016) Updating animal welfare thinking: moving beyond the “Five Freedoms” towards “A Life Worth Living”. Animals, 6(3), pp. 1–20. doi: 10.3390/ani6030021.

Mendl, M., Burman, O. H. P. and Paul, E. S. (2010) An integrative and functional framework for the study of animal emotion and mood. Proceedings of the Royal Society B: Biological Sciences, 277(1696), pp. 2895–2904. doi: 10.1098/rspb.2010.0303.

Miller, L. J. et al. (2020) Behavioral diversity as a potential indicator of positive animal welfare. Animals, 10(7), pp. 1–17. doi: 10.3390/ani10071211.

Millman, S. T. (2007) Sickness behaviour and its relevance to animal welfare assessement at the group level. Animal Welfare, 16(2), pp. 123–125.

Munro, L. A. (2022) Scottish Fish Farm Production Survey 2021. Available at: https://www.gov.scot/publications/scottish-fish-farm-production-survey-2021/pages/5/.

Noble, C., Gismervik, K., Iversen, M. H. et al. (eds.) (2018) Welfare Indicators for farmed Atlantic Salmon: tools for assessing fish welfare http://hdl.handle.net/11250/2575780.

O'Donncha, F. et al. (2021) Data driven insight into fish behaviour and their use for precision aquaculture. Frontiers in Animal Science, 2, pp. 1–18. doi: 10.3389/fanim.2021.695054.

Olden, J. D., Lawler, J. J. and Poff, N. L. (2008) Machine learning methods without tears: A primer for ecologists. The Quarterly Review of Biology, 83(2), pp. 171–193. doi: https://doi.org/10.1086/587826.

Oppedal, F., Dempster, T. and Stien, L. H. (2011) Environmental drivers of Atlantic salmon behaviour in sea-cages: A review. Aquaculture. Elsevier B.V., 311(1–4), pp. 1–18. doi: 10.1016/j.aquaculture.2010.11.020.

Patullo, B. W., Jolley-Rogers, G. and Macmillan, D. L. M. (2007) Video tracking in the extreme : Video analysis for nocturnal. Behavior Research Methods, 39(4), pp. 783–788.

Pinkiewicz, T. H., Purser, G. J. and Williams, R. N. (2011) A computer vision system to analyse the swimming behaviour of farmed fish in commercial aquaculture facilities: A case study using cage-held atlantic salmon. Aquacultural Engineering, 45(1), pp. 20–27. doi: 10.1016/j.aquaeng.2011.05.002.

Polonschii, C., Bratu, D. and Gheorghiu, E. (2013) Appraisal of fish behaviour based on time series of fish positions issued by a 3D array of ultrasound transducers. Aquacultural Engineering, 55, pp. 37–45. doi: 10.1016/j.aquaeng.2013.03.001.

Powell, M. D., Reynolds, P. and Kristensen, T. (2015) Freshwater treatment of amoebic gill disease and sea-lice in seawater salmon production: Considerations of water chemistry and fish welfare in Norway. Aquaculture, 448, pp. 18–28. doi: 10.1016/j.aquaculture.2015.05.027.

Prunet, P. et al. (2012) Fish welfare and genomics. Fish Physiology and Biochemistry, 38, pp. 43–60. doi: 10.1007/s10695-011-9522-z.

Qin, L., Yu, N. and Zhao, D. (2018) Applying the Convolutional Neural Network Deep Learning Technology to Behavioural Recognition in Intelligent Video. Tehnički vjesnik, 25(2), pp. 528–535.

Ravenscraft, E. (2022) The 9 best survey apps for your business, Zapier. Available at: https://zapier.com/blog/best-survey-apps/#SurveyMonkey. Date accessed: 30 June 2023.

Rose, P. and Riley, L. (2019) The use of Qualitative Behavioural Assessment in zoo welfare measurement and animal husbandry change. Journal of Zoo and Aquarium Research, 7(4), pp. 150–161.

RSPCA (2021) RSPCA welfare standards for Farmed Atlantic Salmon, RSPCA welfare standards for Farmed Atlantic Salmon, (February), pp. 1–80. Available at: http://industry.freedomfood.co.uk/media/96636/RSPCA-welfare-standards-for-farmed-Atlantic-salmon-web-version.pdf.

Ruiz-gomez, M. D. L. et al. (2008) Behavioral plasticity in rainbow trout (Oncorhynchus mykiss) with divergent coping styles: When doves become hawks. Hormones and Behaviour, 54, pp. 534–538. doi: 10.1016/j.yhbeh.2008.05.005.

Rushen, J. and De Passille, A. M. B. (2012) Automated monitoring of behavioural-based animal welfare indicators. Animal Welfare, 21, pp. 339–350. doi: 10.7120/09627286.21.3.339.

Saberioon, M. et al. (2017) Application of machine vision systems in aquaculture with emphasis on fish: state-of-the-art and key issues. Reviews in Aquaculture, 9(4), pp. 369–387. doi: 10.1111/raq.12143.

Saberioon, M. M. and Cisar, P. (2016) Automated multiple fish tracking in three-Dimension using a Structured Light Sensor. Computers and Electronics in Agriculture, 121, pp. 215–221. doi: 10.1016/j.compag.2015.12.014.

Santurtun, E., Broom, D. M. and Phillips, C. J. C. (2018) A review of factors affecting the welfare of Atlantic salmon (*Salmo salar*). Animal Welfare, 27(3), pp. 193–204. doi: 10.7120/09627286.27.3.193.

Shortis, M. R. et al. (2016) Progress in the automated identification, measurement and counting of fish in underwater image sequences. Marine Technology Society Journal, 50(1), pp. 4–16.

Sneddon, L. U., Braithwaite, V. A. and Gentle, M. J. (2003) Do fishes have nociceptors? Evidence for the evolution of a vertebrate sensory system. Proceedings of the Royal Society B: Biological Sciences, 270(1520), pp. 1115–1121. doi: 10.1098/rspb.2003.2349.

Southgate, P. and Wall, T. (2001) Welfare of farmed fish at slaughter. In Practice, 23(5), pp. 277–284.

Stien, L. H. et al. (2013) Salmon Welfare Index Model (SWIM 1.0): A semantic model for overall welfare assessment of caged Atlantic salmon: Review of the selected welfare indicators and model presentation. Reviews in Aquaculture, 5(1), pp. 33–57. doi: 10.1111/j.1753-5131.2012.01083.x.

Størkersen, K. V. et al. (2021) Fish protection during fish production. Organizational conditions for fish welfare. Marine Policy, 129, pp. 1–11. doi: 10.1016/j.marpol.2021.104530.

Terayama, K. et al. (2019) Integration of sonar and optical camera images using deep neural network for fish monitoring. Aquacultural Engineering, 86. doi: 10.1016/j.aquaeng.2019.102000.

Tidal -X (2023) Tidal - Protecting the ocean with technology systems while feeding humanity sustainably, Project Tidal. Available at: https://x.company/projects/tidal/. Date accessed: 30 June 2023.

Turnbull, J. F. and Kadri, S. (2007) Safeguarding the many guises of farmed fish welfare. Diseases of Aquatic Organisms, 75(2), pp. 173–182. doi: 10.3354/dao075173.

Valletta, J. J. et al. (2017) Applications of machine learning in animal behaviour studies. Animal Behaviour, 124, pp. 203–220. doi: 10.1016/j.anbehav.2016.12.005.

Vasdal, G. et al. (2022) Qualitative behaviour assessment as part of a welfare assessment in flocks of laying hens. Applied Animal Behaviour Science, 246, pp. 105535. doi: 10.1016/j.applanim.2021.

Veit, W. and Browning, H. (2021) Perspectival pluralism for animal welfare, European Journal for Philosophy of Science. European Journal for Philosophy of Science, 11(1), pp. 1–14. doi: 10.1007/s13194-020-00322-9.

Wang, M. and Takeuchi, H. (2017) Individual recognition and the “face inversion effect” in medaka fish (*Oryzias latipes*). eLife, 6, pp. 1–18. doi: 10.7554/eLife.24728.

Wang, X. (2019) Near-infrared spectroscopy for food quality evaluation. Evaluation Technologies for Food Quality, pp 105-119. doi: 10.1016/B978-0-12-814217-2.00007-X.

Webster, J. (2016) Animal welfare: Freedoms, dominions and “A life worth living”. Animals, 6(6), pp. 2–7. doi: 10.3390/ani6060035.

Wemelsfelder, F. (2007) How animals communicate quality of life: The qualitative assessment of behaviour. Animal Welfare, 16, pp. 25–31. doi: 10.1017/S0962728600031699.

Wemelsfelder, F. and Lawrence, A. B. (2001) Qualitative assessment of animal behaviour as an on-farm welfare-monitoring tool. Acta Agriculturae Scandinavica, Section A - Animal Science, 51(30), pp. 21–25. doi: https://doi.org/10.1080/090647001300004763.

Wiese, T. R. et al. (2023) Concerns and research priorities for Scottish farmed salmon welfare – An industry perspective. Aquaculture, 566, p. 739235. doi: 10.1016/j.aquaculture.2023.739235.

Williams, C. K. I. (2022) Artificial Intelligence for Data Analytics – Final Project Report. London, UK.

Williams, C. K. I. et al. (2022) Artificial intelligence for data analytics (AIDA), The Alan Turing Institute. Available at: https://www.turing.ac.uk/research/research-projects/artificial-intelligence-data-analytics-aida.

Xia, C. et al. (2016) Posture tracking of multiple individual fish for behavioral monitoring with visual sensors. Ecological Informatics, 36, pp. 190–198. doi: 10.1016/j.ecoinf.2016.07.004. Date accessed: 30 June 2023.

Ye, Z. et al. (2016) Behavioural characteristics and statistics-based imaging techniques in the assessment and optimization of tilapia feeding in a recirculating aquaculture system. American Society of Agricultural and Biological Engineers, 59(1), pp. 345–355. doi: doi: 10.13031/trans.59.11406.

Zhao, J., Bao, W., Zhang, F., Zhu, S., Liu, Y., Lu, H., et al. (2018) Modified motion influence map and recurrent neural network-based monitoring of the local unusual behaviors for fish school in intensive aquaculture. Aquaculture, 493, pp. 165–175. doi: 10.1016/j.aquaculture.2018.04.064.

Zion, B. (2012) The use of computer vision technologies in aquaculture – A review. Computers and Electronics in Agriculture, 88, pp. 125–132. doi: 10.1016/j.compag.2012.07.010.

Appendix

Figure A1. Copy of Industry survey (Concerns and research priorities for Scottish farmed salmon welfare)

Determining research priorities for developing on-farm salmon welfare assessment:

When improving salmon welfare, we must first be able to measure and assess salmon welfare.

The GOAL of this survey: 'Determine where research can best improve on-farm welfare measures to be as feasible and effective as possible'.

When assessing welfare, we must cover all welfare concerns. Although health is essential for welfare, there are other ways that welfare can be reduced. This includes:

- Being prevented from performing certain behaviours.
- Being deprived of certain environments from which the animal evolved in.
- Being in a constant state of fear/anxiety (even when the animal is objectively healthy and safe).

When answering these questions, try your best to consider such welfare concerns in addition to the central aspect of maintaining physical health.

Thank you again for your participation!

1. Unique Identification Number *

Enter your answer

2. Current job title: *

Enter your answer

3. Experience in salmon farming (years): *

Enter your answer

4. Please select the following salmon production stages for which you have had any experience of working in: *

- [ ] Hatchery stage
- [ ] Smolt production stage
- [ ] Seawater rearing stage
- [ ] Other

5. Please list any qualifications and/or training that you may have which are relevant to animal welfare:

Enter your answer

Identifying important farming stages & practices, and determining major welfare concerns:

The various stages of salmon farming are all relevant to the fish's welfare, with each stage having unique factors that influence a salmon's quality of life.

6. Please rank the different production stages of a farmed salmon's life-cycle by how much effort should be concentrated towards monitoring and assessing salmon welfare (1 = the most important. A maximum of two different stages may be given the same ranking):

	1	2	3	4	5
Broodstock Stage	○	○	○	○	○
Early Freshwater stage (alevin/fry/parr)	○	○	○	○	○
Smoltification process (change from FW->SW)	○	○	○	○	○
Seawater rearing stage (post-smolts/adults)	○	○	○	○	○
Other (please specify below)	○	○	○	○	○

7. Other production stage (if ranked above):

Enter your answer

8. Comments box (optional): Please feel free to any additional opinions, suggestions or critique with regards to the previous question:

Enter your answer

5007

9. In order of importance, please list a minimum of 3 (up to 5, if possible) of the most significant welfare concerns that you believe currently face farmed salmon; 1st = most important:

Enter your answer

10. Comments box (optional): Please feel free to any additional opinions, suggestions or critique with regards to the previous question:

Enter your answer

11. In order of importance, please list 3 salmon husbandry practices during which are events that require the most attention in monitoring salmon welfare (e.g. crowding / grading / transport / slaughter / treatments / feeding); 1st = most important:

Enter your answer

12. Comments box (optional): Please feel free to any additional opinions, suggestions or critique with regards to the previous question:

Enter your answer

Evaluating welfare indicators by practicality and effectiveness:

IMPORTANT INFORMATION: Some welfare indicators can be developed to provide more information on the welfare status of a fish (e.g. its physical or mental state), while some indicators can be developed to be more practical for on-farm use.

In this next question, you will be asked to rate welfare indicators by these two points:

First point: The PRACTICALITY of the welfare indicators ('how easy are they to use on a farm?').

Second point: The EFFECTIVENESS of the welfare indicators ('what quality of information do they provide on fish welfare, as if they have NO practical limitations at all?').

PRACTICALITY:

Please rate the following welfare indicators, on a scale of 1 to 10, by their practicality for on-farm use (1 = COMPLETELY IMPRACTICAL, 5-6 = SOMEWHAT PRACTICAL, 10 = VERY PRACTICAL).

Under these scores, please list any practical limitations these indicators may currently have. If you are unsure about how to rate a certain indicator, you may skip on to the next one.

13. Practicality score for welfare indicator(s): Grading individuals on their external abnormalities:
Examples include (but are not limited to): Fin/Skin/Eye/Scale condition, wounds or lesions on fish, opercular/vertebral/jaw deformities.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	◯	◯	◯	◯	◯	◯	◯	◯	◯	◯	Very practical

14. Possible limitation(s):

Enter your answer

15. Practicality score for welfare indicator(s): Sampling individuals for physiological measures of stress:

Examples include (but are not limited to): measuring levels of lysozyme, haematocrit, glucose, proteins etc. found within blood/muscle.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

16. Possible limitation(s):

Enter your answer

17. Practicality score for welfare indicator(s): Determining disease/health status of fish by prevalence of certain conditions during routine observations or sampling of individuals:

Examples include (but are not limited to): simple scoring of cataracts, gill bleaching / gill status, scoring for levels of sea lice infestation

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

18. Possible limitation(s):

Enter your answer

19. Practicality score for welfare indicator(s): Presence of acute injuries during husbandry practices:

Examples include (but are not limited to): fin splitting / crush injuries / haemorrhages during hard handling / crowding / pumping.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

20. Possible limitation(s):

Enter your answer

21. Practicality score for welfare indicator(s): Assessing aspects of positive welfare by the presence/absence of enrichment within the production systems:

Examples include (but are not limited to): determining higher positive welfare within hatcheries enriched with artificial kelp, compared to hatcheries devoid of any enrichment.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

22. Possible limitation(s):

Enter your answer

23. Practicality score for welfare indicator(s): Deviations from normal behaviour during routine monitoring:

Examples include (but are not limited to): surface activity, abnormal swimming patterns, increased aggression, decreased feed responses.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

24. Possible limitation(s):

Enter your answer

25. Practicality score for welfare indicator(s): Changes in behaviour during husbandry practices:

Examples of such behaviours include (but are not limited to): signs of panic / exhaustion / disorientation / aggression during crowding / pumping / handling etc.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

26. Possible limitation(s):

Enter your answer

27. Practicality score for welfare indicator(s): Changes in appetite after potentially disturbing husbandry practices:

Gentle reminder: 'Practicality' = How easy this indicator is to measure on a farm-site.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

28. Possible limitation(s):

Enter your answer

5011

29. Practicality score for welfare indicator(s): Production-related parameters:

Examples include (but are not limited to): growth rates, mortality rates, sexual maturation, stage of smoltification etc.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

30. Possible limitation(s):

Enter your answer

31. Practicality score for welfare indicator(s): Duration of time out of water for salmon during certain husbandry practices:

Examples for practices include (but are not limited to): pumping, handling pre-vaccination, and crowding.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

32. Possible limitation(s):

Enter your answer

33. Practicality score for welfare indicator(s): Water quality parameters:

Examples include (but are not limited to): Temperature, Ammonia for FW systems, Harmful Algal Blooms for SW systems, Turbidity etc.

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

34. Possible limitation(s):

Enter your answer

35. Practicality score for welfare indicator(s): Stocking density of rearing system:

	1	2	3	4	5	6	7	8	9	10	
Completely impractical	○	○	○	○	○	○	○	○	○	○	Very practical

36. Possible limitation(s):

5012 Enter your answer

EFFECTIVENESS

Now, assuming that there are no practical limitations involved when using these indicators, please rate each indicator by how effectively they reflect their relevant aspects of salmon welfare (1 = COMPLETELY INEFFECTIVE, 5-6 = SOMEWHAT EFFECTIVE, 10 = VERY EFFECTIVE):

For example: "How effectively, from 1 - 10, does fin condition reflect the physical condition of a salmon?". If you are unsure about how to rate a certain indicator, you may skip on to the next one.

38. Effectiveness score for welfare indicator(s): Grading individuals on their external abnormalities:

Examples include (but are not limited to): Fin/Skin/Eye/Scale condition, wounds or lesions on fish, opercular/vertebral/jaw deformities etc.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

39. Effectiveness score for welfare indicator(s): Sampling individuals for physiological measures of stress:

Examples include (but are not limited to): measuring levels of lysozyme, haematocrit, glucose, proteins etc. found within blood/muscle.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

40. Effectiveness score for welfare indicator(s): Determining disease/health status of fish by prevalence of certain conditions during routine observations or sampling of individuals:

Examples include (but are not limited to): simple scoring of cataracts, gill bleaching / gill status, scoring for levels of sea lice infestation.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

41. Effectiveness score for welfare indicator(s): Presence of acute injuries during husbandry practices:

Examples include (but are not limited to): fin splitting / crush injuries / haemorrhages during hard handling / crowding / pumping.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

42. Effectiveness score for welfare indicator(s): Assessing aspects of positive welfare by the presence/absence of enrichment within the production systems:

Examples include (but are not limited to): determining higher positive welfare within hatcheries enrich with artificial kelp compared to hatcheries devoid of any enrichment

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

5013

43. Effectiveness score for welfare indicator(s): Deviations from normal behaviour during routine monitoring:

5014 *Examples include (but are not limited to): surface activity, abnormal swimming patterns, increased aggression, decreased feed responses).*

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

44. Effectiveness score for welfare indicator(s): Changes in behaviour during husbandry practices:

Examples of such behaviours include (but are not limited to): signs of panic / exhaustion / disorientation / aggression during crowding / pumping / handling etc.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

45. Effectiveness score for welfare indicator(s): Changes in appetite after potentially disturbing husbandry practices:

Gentle reminder: These scores are simply for how well these measures reflect fish welfare, assuming NO practical limitations are involved

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

46. Effectiveness score for welfare indicator(s): Production-related parameters:

Examples include (but are not limited to): growth rates, mortality rates, sexual maturation, stage of smoltification etc.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

47. Effectiveness score for welfare indicator(s): Duration of time out of water for salmon during certain husbandry practices:

Examples of husbandry practices include (but are not limited to): pumping, handling pre-vaccination, crowding.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

48. Effectiveness score for welfare indicator(s): Water quality parameters:

Examples include (but are not limited to): Temperature, Ammonia for FW systems, Harmful Algal Blooms for SW systems, Turbidity etc.

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

49. Effectiveness score for welfare indicator(s): Stocking density of rearing system:

	1	2	3	4	5	6	7	8	9	10	
Completely ineffective	○	○	○	○	○	○	○	○	○	○	Very effective

Research priorities for developing welfare indicators:

This next section will ask you to rate the RELEVANCE and URGENCY behind improving certain welfare indicators in different ways.

Now, on a similar scale, please rate the urgency of developing these welfare measures:

1 = NOT URGENT AT ALL, 5-6 = SOMEWHAT URGENT, 10 = EXTREMELY URGENT.

If you are unsure about how to rate one of the following research outcomes, you may skip on to the next one.

57. Developing understanding behind environmental conditions (e.g. optimal light conditions, turbidity, & total suspended solids for each specific life stage in salmon (parr, smolts & post-smolts)

This could help ensure that the quality of early life stages are not jeopardised, and that later quality of life is not affected through improper development.

Not urgent at all 1 2 3 4 5 6 7 8 9 10 Extremely urgent

58. Developing more fish/user-friendly methods for welfare indicators which currently require catching & handling of the fish (e.g. having to sample cages for scoring physical injury, body condition, malformations):

These processes still have potential in disturbing salmon during the capture process

Not urgent at all 1 2 3 4 5 6 7 8 9 10 Extremely urgent

Completely irrelevant Extremely relevant

59. Developing the ability to quantify fish behaviours with monitoring systems (e.g. passive, vision-based / acoustic devices):

Developing such systems more towards quantifying certain fish behaviours could allow for a more detailed analysis of welfare through behavioural indicators.

Not urgent at all 1 2 3 4 5 6 7 8 9 10 Extremely urgent

60. Developing welfare indicators that are currently only able to be carried out in the lab, to the point where they can become operational on farm sites:

Such indicators could provide a closer insight to the welfare of the animals that otherwise could only have been done within a laboratory setting.

Not urgent at all 1 2 3 4 5 6 7 8 9 10 Extremely urgent

61. Developing welfare indicators that allow for the remote monitoring of the salmon:

These indicators could help with the safeguarding of salmon welfare when access for staff to the farm sites becomes limited (e.g. during storms for sea cages, or during pandemics which limit staff presence).

Not urgent at all 1 2 3 4 5 6 7 8 9 10 Extremely urgent

Determining when welfare monitoring/assessment best fits within the farmer's routine

For any welfare assessment to be effective, the welfare indicators must be used in such a way that best falls within the farm staff's routines, thus minimising any conflicts with their other responsibilities.

63. Which parts of a farmer's routine (daily, or during specific tasks) do you believe provide the most suitable opportunities for monitoring certain welfare indicators with the salmon (please select a maximum of THREE options):

- ☐ During feeding times
- ☐ During health checks
- ☐ During routine cage/tank inspections
- ☐ During grading and/or transfer
- ☐ During video monitoring
- ☐ Other

64. Comments box (optional): Please feel free to any additional opinions, suggestions or critique with regards to the previous question:

Enter your answer

END OF SURVEY

5017

Table A1. Grouping of responses into categories of overall welfare concerns

Total weighted score, in terms of importance, for each category in Section 2 (Overall welfare concerns):		
Category:	**Total Score:**	**Responses included:**
Sea lice	155	"Sea lice"
Gill health	120	"Gill health", "Gill problems", "AGD or PGD", "Gill Disease", "Gill health challenges"
Environmental challenges	115	"Poor oxygen", "Tidal throughput", "High energy sites", "Rising sea temperatures", "Climate change", "Plankton / algal blooms", "Jellyfish", "Reliability of RASs & WQ of systems", "Background microbiology"
Interventions	74	"Too many treatments", "Invasive treatments", "Mechanical / non-medicinal sea lice treatments", "Vaccinations / treatments", "Improper treatments", "Overcrowding during treatments"
Handling	65	"(Im)proper handling", "Handling damage", "Harvesting", "Grading", "Increased handling due to health challenges", "Handling for transport"
Non-gill health issues	60	"Diseases", "PD", "Anaemia", "Health Issues", "Viral/Bacterial diseases (PMCV, PRV, SAV)", "CMS", "RTFS", "Disease Challenges", "Haemorrhagic Smolt Syndrome", "Spread of disease", "Fin & Skin condition (lesions)"
Stocking density	51	"SD", "Stocking Density", "High SD"
Staff training	33	"Poor decision making in cluster sites", "Lack of training / knowledge", "Poor decision making during treatments / handling" , "Respect / care when working with fish", "Neglect", "Under feeding", "Lack of stimulation"
Predation	22	"Predators (Seals)". "Predation (Control)"
Farm management	21	"Company strategies", "Commercial pressure", "Management of environment", "Increasing production with no focus on individual health", "Senior Mgmt. focusing on profit", "Lack of focus on animals (treated as numbers)", "No adaptation to previous health issues", "Regulatory pressures (drive for compliance to set thresholds"

Table A2. Grouping of responses into categories of husbandry practices

Total weighted score, in terms of importance, for each category in Section 3 (Husbandry practices):		
Category:	**Total Score:**	**Responses included:**
Interventions	121	"Treatments", "Vaccinations", "Mechanical / chemical / bath / medicinal / delousing treatments", "Enclosed interventions"
Handling	120	"Crowding", "Grading", "(Physical) handling", "Crowding for treatments", "Post-treatment handling"
Feeding	34	"Feeding", "First feeding"
Transport	20	“Transport”
Slaughter	19	“Slaughter”
Smolt transfers	17	"Smolt transfers", "Loading smolts from FW to wellboat", "Discharge of smolts from wellboat to SW", "Smoltification of population"