

REVIEW

Applying genetic technologies to combat infectious diseases in aquaculture

Nicholas A. Robinson^{1,2}  | Diego Robledo³  | Lene Sveen¹  |
 Rose Ruiz Daniels³  | Aleksei Krasnov¹ | Andrew Coates²  | Ye Hwa Jin³ |
 Luke T. Barrett^{2,4}  | Marie Lillehammer¹  | Anne H. Kettunen¹  |
 Ben L. Phillips² | Tim Dempster²  | Andrea Doeschl-Wilson³  |
 Francisca Samsing⁵  | Gareth Difford¹ | Sarah Salisbury³ | Bjarne Gjerde¹ |
 John-Erik Haugen¹ | Erik Burgerhout¹  | Binyam S. Dagnachew¹  |
 Dominic Kurian³ | Mark D. Fast⁶ | Morten Rye⁷ | Marcela Salazar⁷ |
 James E. Bron⁸  | Sean J. Monaghan⁸  | Celeste Jacq⁹ | Mike Birkett¹⁰ |
 Howard I. Browman¹¹ | Anne Berit Skiftesvik¹¹ | David M. Fields¹² |
 Erik Selander¹³ | Samantha Bui⁴  | Anna Sonesson¹ | Stanko Skuger¹⁴ |
 Tone-Kari Knutsdatter Østbye¹ | Ross D. Houston⁷ 

¹Nofima AS, Tromsø, Norway

²Sustainable Aquaculture Laboratory—Temperate and Tropical (SALTT), School of BioSciences, The University of Melbourne, Melbourne, Victoria, Australia

³The Roslin Institute and Royal (Dick) School of Veterinary Studies, The University of Edinburgh, Edinburgh, UK

⁴Institute of Marine Research, Matre Research Station, Matredal, Norway

⁵Sydney School of Veterinary Science, The University of Sydney, Camden, Australia

⁶Atlantic Veterinary College, The University of Prince Edward Island, Charlottetown, Prince Edward Island, Canada

⁷Benchmark Genetics, Bergen, Norway

⁸Institute of Aquaculture, University of Stirling, Stirling, Scotland, UK

⁹Blue Analytics, Kong Christian Frederiks Plass 3, Bergen, Norway

¹⁰Rothamsted Research, Hertfordshire, UK

¹¹Institute of Marine Research, Austevoll Research Station, Ecosystem Acoustics Group, Tromsø, Norway

¹²Bigelow Laboratory for Ocean Sciences, Boothbay, Maine, USA

¹³Department of Marine Sciences, University of Gothenburg, Gothenburg, Sweden

¹⁴Cargill Aqua Nutrition, Bergen, Norway

Correspondence

Nicholas A. Robinson, Hutchinson Avenue,
Warrandyte, VIC 3113, Australia.
Email: nick.robinson@nofima.no

Funding information

Biotechnology and Biological Sciences
Research Council, Grant/Award Number: BB/
V009818/1; Fiskeri - og havbruksnæringens
forskningsfond, Grant/Award Number:

Abstract

Disease and parasitism cause major welfare, environmental and economic concerns for global aquaculture. In this review, we examine the status and potential of technologies that exploit genetic variation in host resistance to tackle this problem. We argue that there is an urgent need to improve understanding of the genetic mechanisms involved, leading to the development of tools that can be applied to boost host

This is an open access article under the terms of the [Creative Commons Attribution](https://creativecommons.org/licenses/by/4.0/) License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

© 2022 The Authors. *Reviews in Aquaculture* published by John Wiley & Sons Australia, Ltd.

901631; Norges Forskningsråd, Grant/Award Numbers: 244131, 320619

resistance and reduce the disease burden. We draw on two pressing global disease problems as case studies—sea lice infestations in salmonids and white spot syndrome in shrimp. We review how the latest genetic technologies can be capitalised upon to determine the mechanisms underlying inter- and intra-species variation in pathogen/parasite resistance, and how the derived knowledge could be applied to boost disease resistance using selective breeding, gene editing and/or with targeted feed treatments and vaccines. Gene editing brings novel opportunities, but also implementation and dissemination challenges, and necessitates new protocols to integrate the technology into aquaculture breeding programmes. There is also an ongoing need to minimise risks of disease agents evolving to overcome genetic improvements to host resistance, and insights from epidemiological and evolutionary models of pathogen infestation in wild and cultured host populations are explored. Ethical issues around the different approaches for achieving genetic resistance are discussed. Application of genetic technologies and approaches has potential to improve fundamental knowledge of mechanisms affecting genetic resistance and provide effective pathways for implementation that could lead to more resistant aquaculture stocks, transforming global aquaculture.

KEYWORDS

gene editing, genomic selection, host resistance, sea lice, transcriptomics, white-spot syndrome virus

1 | INTRODUCTION

'Blue foods' have been highlighted as a major source of nutrients supporting the health and livelihood of many communities throughout the world.¹ Aquaculture, currently the fastest growing food production industry, plays a key role in producing and supplying fish, shellfish and algae. However, the mass culture of animals in any environment carries with it a high risk of contracting, propagating and spreading infectious disease.^{2,3} Some diseases affecting fish and shellfish can lead to 100% mortality or necessitate complete destocking. Disease prevention and treatment are necessary, but current options are often costly, ineffective and can negatively impact animal welfare, local ecosystems and product quality. For example, biosecurity is particularly challenging when animals are farmed in an open water system, and logistical difficulties in handling makes it challenging to vaccinate and treat individual animals. Here, we focus mainly on the improvement of host disease resistance which can be defined as the host's ability to reduce pathogen invasion (i.e., limiting pathogen entry into target tissues and replication).

Vaccination has proven to be an effective preventative measure boosting immunity for many major diseases affecting humans (e.g.,⁴), domestic animals and livestock including fish.^{5–12} In some instances, total eradication of human or livestock disease has been possible through the implementation of vaccination programmes.^{13,14} However, vaccines are difficult to develop against some diseases, especially against ectoparasites such as sea lice (crustacean copepods) or amoebae.¹⁵ Moreover, vaccination has limited success for host

species with a less developed adaptive immune system such as crustaceans or molluscs.¹⁶ Additional tools for improving host resistance are therefore needed. In this review, we describe the use of genetic technologies that can exploit and potentially also create genetic variation in disease resistance. Such approaches can both provide a greater understanding of the natural mechanisms affecting disease resistance and can be used to boost host resistance and thereby reduce the impact of infectious diseases in aquaculture. The high fecundity of aquatic species leads to opportunities for rapid propagation and dissemination of strains with improved innate disease resistance.

Two of the most valuable global aquaculture species are salmon (with 2.6 million tonnes yearly production of Atlantic salmon, *Salmo salar*, valued at US\$ 17 billion in 2019) and shrimp (with 6.2 million ton of both whiteleg shrimp *Litopenaeus vannamei* and black tiger shrimp *Penaeus monodon* valued at US\$ 38.5 billion in 2019).¹⁷ The intensification and global nature of both Atlantic salmon and shrimp farming have led to major challenges in the form of infectious diseases. Currently, sea lice in Atlantic salmon and white spot syndrome in shrimp are two of the most pressing problems for these aquaculture industries.

Sea lice (Caligidae) are marine ectoparasitic copepods that attach to host organisms and feed on mucus, skin and blood. Most lice species have a free-swimming planktonic larval phase (copepodid) that facilitates host-finding, and once attached to a host, they undergo two further moults through sessile (immobile) stages before reaching mobile pre-adult stages that can move around the host and exhibit host-switching behaviours. They then become adults, reproducing sexually on the same host and releasing fertilised eggs into the water

column.^{18–20} Sea lice have been found on numerous species of wild and farmed fish, and while the prevalence may be high, infestation densities are usually low and have relatively minor impacts on production or animal welfare (e.g.,^{21–23}). The Atlantic salmon farming industry is one notable exception in which infestations can be severe and lead to poor welfare and mortality for farmed fish.^{24–26} Strict delousing regimes are used to keep lice infestation intensities low, but some delousing regimes (involving crowding, pumping and lice removal effects) can pose severe welfare risks.^{27–29} The lice larvae produced on-farm can also infest wild salmonid populations.^{30–35} Sea lice cost the Norwegian salmon sector at least 7.3 billion kroner (~US\$ 810 million) per annum³⁶ (up from US\$ 435 million estimated for 2011³⁷) and are of major concern for other salmon industries around the world. The welfare and economic impacts of louse parasitism are predicted to be even worse with higher seawater temperatures.^{38–40} *Lepeophtheirus salmonis* is the louse species currently of greatest concern in Norway, but *Caligus elongatus* has become an increasing problem (particularly in northern latitudes,⁴¹) and *Caligus rogercresseyi* is of great concern in Chile.^{42–44} Lice control measures entail substantial costs,⁴⁵ with no single method proving completely effective,⁴⁶ and moreover, lice have already evolved resistance to some methods of control (reviewed in Reference [47] e.g.⁴⁸). Repeated reintroduction of high-density naïve salmon hosts to sea cages is effectively a serial passage experiment that disproportionately favours the parasite's adaptation to treatments.⁴⁹ There is an urgent need to develop new strategies to mitigate the impact of sea louse parasitism.

White spot syndrome virus (WSSV) causes devastating disease that severely impacts global shrimp aquaculture.⁵⁰ All decapod crustaceans, including all major cultured shrimp species, are affected by WSSV, and disease outbreaks typically cause mass mortality within a few days.⁵¹ Elimination of the virus from the open pond systems in which shrimp are extensively grown has not been possible.⁵² Despite the lack of an adaptive immune system, stimulation of the innate immune system of shrimp by immune priming shows promise but has not yet been proven to prevent and control disease in the field.^{53–55}

These two infectious diseases have a large impact on these aquaculture sectors, but similar disease problems affect all aquaculture species with devastating consequences.^{56,57} Alternative measures for preventing infestation and infection are needed. Modern genetic and genomic technologies and methodologies can help us understand why some individuals, strains and species have higher inherent abilities to resist certain diseases. Such knowledge can be applied to boost the ability of farmed fish and shellfish to resist disease and reduce the burden of infectious diseases in aquaculture. Here, we will review the recent application of these technologies and explore their potential to improve the resistance of aquaculture stocks to infectious diseases in the near future.

2 | IMPROVING HOST RESISTANCE AS A PREVENTATIVE MEASURE

Genetic improvement by selective breeding is playing a crucial role in helping aquaculture meet future demands for animal protein,⁵⁸ and

research into the genetics of disease resistance has played a major role in ensuring the health and security of aquaculture stocks.⁵⁹ The level of host susceptibility or resistance is usually influenced by many genes each contributing small additive effects (also known as a polygenic trait). Disease resistance is usually heritable,⁵⁹ with moderate to low heritability estimates for resistance to sea lice in Atlantic salmon (0.3⁶⁰; 0.26–0.33⁶¹; 0.02–0.14⁶²; 0.22⁶³; 0.14–0.43⁶⁴; 0.28⁶⁵; 0.22–0.33⁶⁶) and WSSV in shrimp (0.01–0.31,^{67–71}). The potential for improving lice resistance through selective breeding is substantial as the coefficient of variation in lice count per fish is very high ranging from 60.5 to 95.0.⁶¹ These are within-population estimates of heritability, but there are also very clear differences across species in susceptibility to parasitism and disease. For example, some salmonid species (e.g., coho *Oncorhynchus kisutch* and pink *Oncorhynchus gorbuscha*) have a higher innate ability to resist sea lice infection than others (e.g., Atlantic salmon *Salmo salar*, chum *Oncorhynchus keta* and rainbow trout *Oncorhynchus mykiss*).^{72–75} Thus, there is genetic variation for resistance both within and between species that can be exploited for tackling this infectious disease.

For lice resistance, a key breeding objective trait of importance is the number of lice treatments in a production cycle (or per year) at a farm. This is determined by the treatment threshold used (the maximum number of adult female lice per fish), intensity of lice infestation and the host resistance to sea lice. A strong treatment threshold may introduce more frequent lice treatments and thus higher costs. Due to rules requiring a low threshold for delousing, it is not likely that a reduction in the number of lice treatments can be observed at a farm level over a few generations of selection. Therefore, breeding companies are reluctant to put too much weight on the lice resistance trait as this will result in reduced genetic gain for the other traits selected that pay off within a shorter time horizon. However, such a reduction is expected to be seen at a company, and in particular, at a national level. Furthermore, as direct selection for reduced number of lice treatments is not possible to apply, indirect selection through reduced lice counts per fish, or lice density obtained from a controlled challenge test, must be used instead, but this is often less efficient than direct selection.

We also need to be sure that the selection or intervention applied adequately covers interactions occurring at all relevant life stages of the pathogen and the host. In the case of salmon lice, the genetic correlation between the number of attached and adult lice counted at different times has been found to be high,⁶² and there is also a documented high genetic correlation between the number of lice per fish recorded at two subsequent challenge tests with de-lousing after the first,⁶⁴ as well as a high genetic correlation between the number of lice recorded on the same fish in two different times of the year (Gjerde, Pers. comm.). These results show that selection for increased resistance to lice based on lice count per fish or lice density will provide increased resistance to relevant life stages of the lice at relevant life stages of the host.

The long-term objective of selection for increased pathogen resistance is to develop a host that the pathogen does not infect under field conditions so that the pathogen will not be able to reproduce

and spread in farmed and close by wild host populations. Genetic correlations between disease resistance in a challenge test tank and under conditions in the field vary depending on the disease and are influenced by many different environmental factors. High correlations have been reported for host resistance to sea lice⁶² and to furunculosis caused by the bacterium *Aeromonas salmonicida*,⁷⁶ and they are also likely high for host resistance to WSSV (near identical genetic ranking for survival rate among susceptible and resistant lines,⁷⁷). This is encouraging as selection for WSSV or sea lice host resistance based on controlled challenge test data should benefit the farmers that utilise selected seedstock. On the other hand, low genetic correlations between tank and field challenges have been detected for some other parasitic diseases (e.g., amoebic gill disease in Atlantic salmon⁷⁸).

Major challenges to genetic improvement of host disease resistance could be caused by trade-offs between resistance, transmissibility and growth. For instance, in shrimp, there is a clear negative and thus unfavourable genetic correlation between resistance to WSSV and growth rate (-0.55 to -0.64 ,^{69,71}). In Atlantic salmon, the genetic correlation between resistance to sea lice and growth rate (-0.32 to -0.37) is also reported to be unfavourable⁶² implying that both traits need to be recorded and selected to obtain desired genetic gains for both growth rate and sea lice resistance. Another important issue that is seldom considered when selecting for improved disease resistance is possible trade-offs between host resistance, tolerance and infectivity. Animals with high tolerance could survive longer and thus be able to transmit infectious pathogens for longer.⁷⁹ In such instances, genetic improvement of host tolerance might not lead to beneficial epidemiological effects.

To make genetic gains in disease resistance, it would be desirable to breed from survivors to a disease if it were not possible for the survivors to transmit and propagate the disease further in the host population (i.e., hosts surviving WSSV can transmit disease). In such cases when transmission and propagation of the disease by survivors is a risk, selective breeding programmes are restricted to selection of uninfected breeding candidates (kept in a bio-secure breeding nucleus). In the past, this selection has been based on the average performance of the infected siblings to the breeding candidates and it was not possible to capture the within-family or Mendelian sampling component of genetic variation for sib recorded traits. Developments in genomics have enabled quantitative geneticists to exploit the broad genetic variation for host disease resistance that exists within families in these circumstances.

Marker-assisted selection was initially touted as a technology of great promise for genetic improvement. However, because most disease traits measured to date are polygenic, and therefore single genetic markers (or single quantitative trait loci [QTL]) generally account for a small fraction of the overall genetic variation, there are few examples where the application of marker-assisted selection has resulted in realised genetic gains larger than expected when using traditional pedigree-based selective breeding. A notable exception where marker-assisted selection has had a demonstrable large benefit in aquatic species is in resistance to infectious pancreatic necrosis in Atlantic salmon, where a single gene accounts for a large proportion of the overall genetic variation.^{80–82} Nonetheless, while the potential

value of genomic intervention needs to be assessed for each target trait and culture environment, the latest methodologies for utilising genomic information (i.e., genomic selection,⁸³) have been predicted to have high potential benefit for disease resistance and other traits that cannot be recorded on the breeding candidates,⁸⁴ many of which are known to be polygenic in nature and difficult to directly assess on unexposed animals.

Finally, recent developments in genomic technologies provide opportunities to better understand the genetic mechanisms affecting the differences in host resistance between and within species. Not only can these technologies improve our understanding of the mechanisms involved, but they can help develop tools that could be used to reduce the impact of infectious diseases. For instance, using clustered regularly interspaced short palindromic repeat (CRISPR-Cas) genome editing technology provides a way for making targeted changes to genes found to influence host resistance.⁸⁵ However, the implementation of these technologies, and dissemination of genetically improved fish and shellfish for production by the industry, needs to be carefully considered to avoid potential negative environmental consequences (e.g., escapees interbreeding with wild counterparts) and counter-adaptation by disease agents. All of these issues need to be addressed to devise an ethical and sustainable long-term strategy for eliminating or reducing pathogen infection. As farmed hosts such as Atlantic salmon can be a major propagator, and hence, driver of disease,³⁴ such a strategy should also have long-term benefits in providing a form of 'herd immunity' for naturally existing populations of host species that inhabit areas close to major aquaculture environments.

3 | BIOLOGICAL MECHANISMS AFFORDING HOST RESISTANCE

Host resistance to infection and parasitism is highly complex and most likely determined by a range of responses (Figure 1), and indeed research across a range of fish species has identified broad potential mechanisms affecting host resistance.⁸⁶ Comparatively less is known about the biology of resistance to disease in invertebrates (e.g., shrimp, Figure 2).

3.1 | Immune and cellular response to infection and infestation

The innate immune system provides a pivotal first defence against invading pathogens and exists across invertebrate and vertebrate organisms (e.g., proposed innate immune response to WSSV in shrimp, Figure 2). Most organisms possess cellular receptors, which bind to foreign elements and differentiate self from non-self. These pattern recognition receptors (PRRs) recognise pathogen-associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs). There are several classes of PRRs, including Toll-like receptors (TLRs), NOD-like receptors (NLRs) and RIG-I-like receptors (RLRs), each recognising distinct microbial components and directly

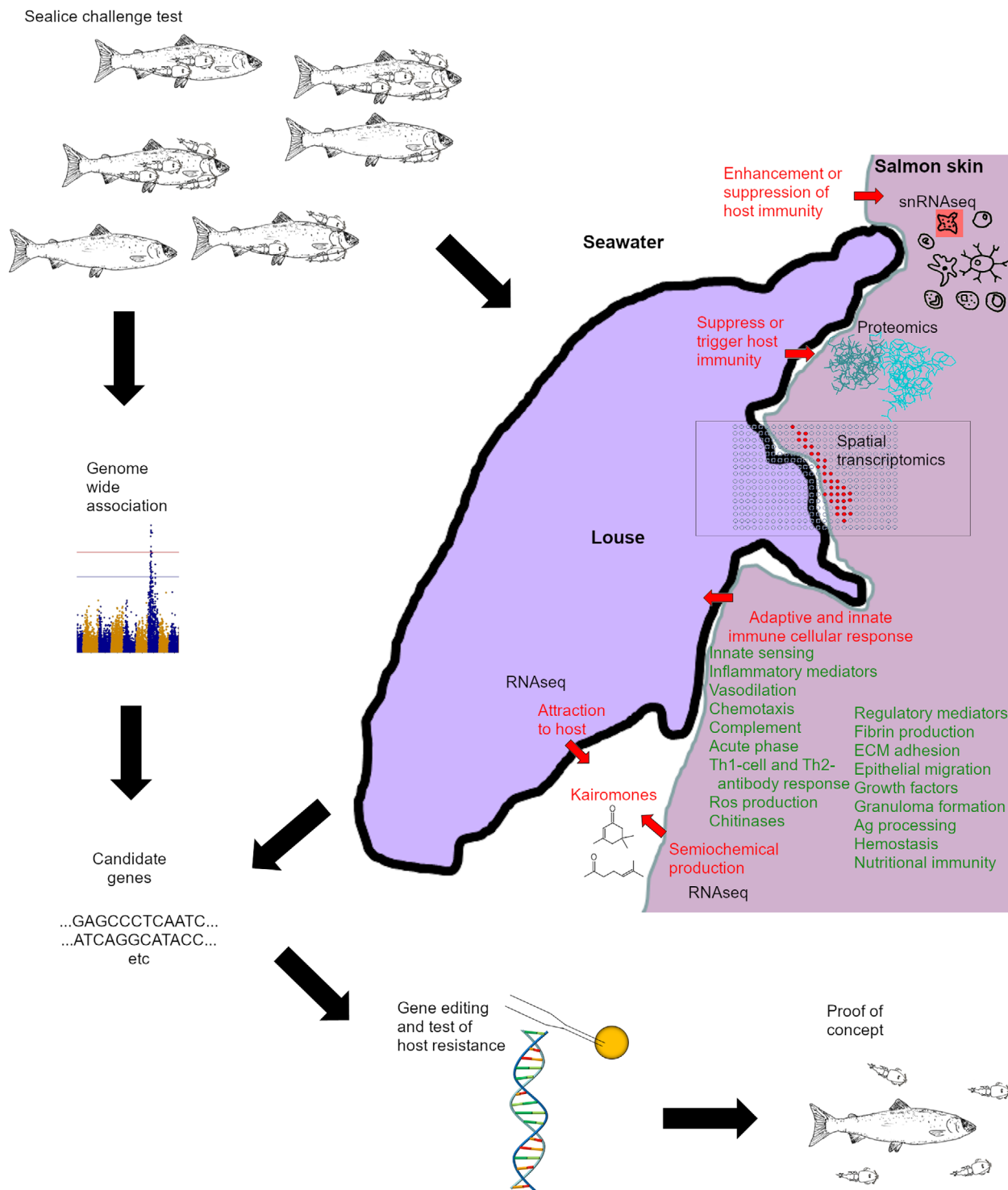


FIGURE 1 Exploring the genetic basis of mechanisms providing host resistance. Host resistance to sea lice is likely affected by environmental and dietary factors that enhance or suppress salmon immunity, the immune cellular response (adaptive and innate immune systems), kairomones that attract the lice to the host and proteins that are secreted by the louse and suppress or trigger host immunity (red text). More detailed processes and factors likely to promote host immunity in coho, pink and more resistant strains of Atlantic salmon are listed in green text. To search for genes in the host that are up- or down-regulated at key time points post-infection: (1) genome-wide association studies can identify genes mapping to chromosomal regions associated with host resistance, (2) single nuclei RNAseq (snRNAseq) can be used to study which populations of cell types are responding in host tissue close to the interface between the salmon and the louse, (3) spatial transcriptomics and spatial proteomics can be used to map precisely where the response occurs, (4) proteomics can be used to discover interactions between host cell and lice immunomodulatory proteins (suppressing or triggering host immunity), (5) RNAseq can be used to study semiochemical production by the host and transcriptomic response of the louse in response to kairomones, and, (6) gene editing can be used to test putative genes affecting host resistance, by experimentally challenging edited and non-edited salmon with sea lice and comparing counts of attached lice

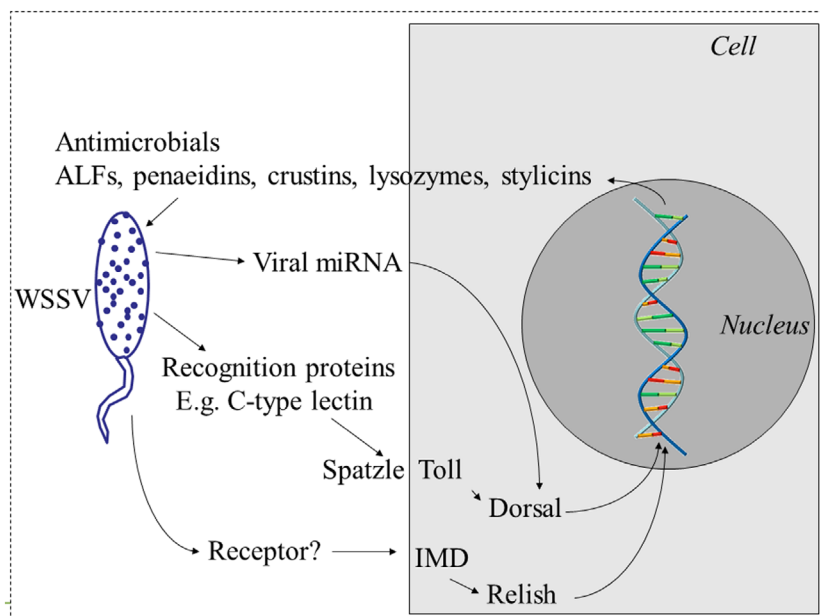


FIGURE 2 Simplified diagrammatic representation of the proposed immune response to white spot syndrome virus (WSSV) in shrimp. WSSV enters the cell using endocytic routes and induces both humoral and cellular responses from the host. Toll and immune deficiency (IMD) pathways are activated and dorsal and relish are translocated to the nucleus inducing the expression of antimicrobial peptides that limit viral replication. C-type lectin proteins can neutralise WSSV by binding to envelope proteins. Interestingly, WSSV is able to hijack the humoral pathways in order to facilitate its replication

activating immune effector cells. Whether hemocytes of crustacea, or macrophage/dendritic/B-lymphocytes of teleosts, these phagocytic cells eliminate potential pathogens and act as the bridge to adaptive immune activation in vertebrates.

Large differences in the louse settlement success, host immune response and consequent infection of different salmon species are observed after exposure to sea lice.⁷³ Although counts of settled copepodids can be initially similar in Atlantic, chum, coho, pink salmon and rainbow trout in experimental challenge tests, the intensity of infection drops in the resistant Pacific species in the first days post-challenge.⁷² Coho and pink salmon mount a more effective response in the skin where the lice attach than Atlantic salmon, despite rapid and large-scale immune gene activation in the skin of Atlantic salmon,^{65,87–93} suggesting that resistance is determined by the character rather than magnitude of response. Resistance to the parasite seems a highly complex biological process. Within 24 h of parasite settlement coho salmon exhibit a multifocal inflammatory granulocytic infiltration, associated with a pronounced non-specific epithelial hyperplasia and melanin deposition over the coming days. Epidermal hyperplasia is intensified and accompanied by eosinophilic granular cell infiltration by 7 days and encapsulation of the parasite by 10 days post infestation, with limited pathology thereafter.⁹⁴

In contrast, this response is not observed in pink salmon (also highly resistant), which instead develop a rapid inflammatory response.^{74,95} Tadiso et al.⁹³ found transient increases of mannose-binding receptors and C-type lectins in Atlantic salmon skin, yet these were minor and largely absent after 3 days post-infection. In coho salmon, local and systemic induction of chitin, mannose and other

non-self PRRs initiate the acute inflammation preceding louse encapsulation and rejection. Similarly, whereas acute phase response genes, extracellular matrix (ECM) and tissue remodelling matrix metalloproteinases (MMPs) are all down-regulated in this time frame for Atlantic salmon, they are upregulated in Coho salmon.^{96–98}

Although Atlantic salmon develops a rapid local and systemic transcriptomic response to *L. salmonis*, this does not result in substantial levels of protection in most individuals,⁹³ and immunomodulation by the parasite has been suggested to play an important role in the early stages of louse attachment.^{74,99–106} A range of anti-host proteins, including proteolytic enzymes and prostanoids, are secreted by a variety of louse exocrine glands and pass to host tissues from the point of louse attachment. These serve to suppress host immune responses during the initial stages of attachment^{107–109} and different protease profiles have been identified between excretory/secretory (E/S) products from pre-adult and adult *L. salmonis*.¹¹⁰ Host- and context-specific expression of genes in the louse (e.g.,^{111–113}) may also provide a key to understanding species-specific levels of host resistance.

Similarly, infection with WSSV in shrimp results in a complex range of host-pathogen interactions in humoral and cellular pathways (Figure 2). The virus may enter the cell by several endocytic routes, and after this Toll and immune deficiency (IMD) signalling pathways are activated resulting in the up-regulation of the NF- κ B transcription factors Dorsal, Relish and AP-1 that increase the expression of antimicrobial peptides (AMP).^{114,115} To date, several classes of AMPs or effectors have been identified in shrimp: Penaeidins (PEN), Crustins (Cru), anti-LPS-factors (ALF), C-type lectins (CTL), Lysozymes (Lyz), and thioester-containing proteins (TEP).^{116,117}

WSSV has also been shown to immunomodulate the shrimp response. Inhibition of the NF- κ B signalling is achieved by encoded microRNAs (WSSV-miR-N13 and WSSV-miR-N23), which can target Dorsal.¹¹⁸ WSSV also modulates the Toll dorsal pathway at different levels, for example, the WSSV449 protein; homologous to tube insect protein, can activate NF- κ B to trigger promoters of viral genes such as *wsv069*, *wsv303* and *wsv371*.¹¹⁹

Another pathway relevant to the immune responses of both invertebrates and vertebrates is the JAK/STAT pathway. In shrimp, activation of the JAK/STAT pathway by bacterial challenge increases expression of *AMPS ALF-A1*, *ALF-C1*, *ALF-C2*, *Cru1-1* and *Cru1-5*.¹²⁰ Upon WSSV infection in shrimp the expression of phosphorylated STAT is upregulated, and STAT is translocated to the nucleus. However, the binding motif of STAT binds to the WSSV 1E1 promoter region facilitating, instead of inhibiting, viral replication.¹²¹

Apoptosis also plays a critical role in vertebrate and invertebrate defence against viral pathogens. In insects, apoptosis has an antiviral action to control viral pathogens,¹²² in shrimps, however, the role of apoptosis during WSSV infection is not clear. The viral accommodation theory says that apoptosis can be used by the virus to promote infection and that reduced rates of cell death can be the cause of the reduction of viral pathogenicity in shrimps.¹²³ However, other reports have shown that cells displaying nuclear condensation and fragmentation characteristics of apoptosis did not contain WSSV virions, whereas those containing WSSV virions were not apoptotic,^{124,125} suggesting that WSSV is able to inhibit apoptosis of infected cells. Several mechanisms have been identified, with caspases as the main target of viral proteins. The viral protein AAP-1, encoded by WSSV449, contains two putative caspase-9 cleavage sites, VETD233G and LEHD303G, as well as a caspase-3 cleavage site, DEVD272G. WSV222, which is an E3 ubiquitin ligase, blocks apoptosis through the ubiquitin-mediated degradation of the shrimp pro-apoptotic TSL protein.¹²⁶

A holistic approach using 'omic' technologies (including metabolomics¹²⁷ and glycomics¹²⁸ which are not detailed in this review) could help elucidate the complex interaction between WSSV and the crustacean host. WSSV has been shown not only to modulate the hosts immune response, but also to interfere with shrimp cell metabolism, being the first invertebrate virus known to induce aerobic glycolysis in infected cells (Warburg effect)¹²⁹ via the PI3K-Akt- mTOR pathway, which is activated upon WSSV infection. Metabolic reprogramming induced by WSSV involves glycolysis, the tricarboxylic acid (TCA) cycle, glutaminolysis and lipid metabolism¹³⁰ (Figure 3). Su et al.¹³¹ demonstrated that pre-treatment of shrimp with a PI3K inhibitor was associated with a decrease in WSSV gene expression and copy number, indicating the WSSV-induced Warburg effect is important for successful viral replication. Hypoxia-inducible factor-1 (HIF-1), a transcriptional factor that regulates the expression of several glycolytic genes, is also involved in the metabolic reprogramming induced by WSSV. HIF-1 silencing in WSSV-infected shrimps blocks the upregulation of the glycolytic enzymes HK, PFK and PK and the expression and activity of LDH, increasing survival of the infected animals.¹³²

Immune training or priming using heat-killed pathogens, heat shock proteins or poly(I:C) mimicking double-stranded viral RNA can

trigger strong effector immune responses in invertebrates^{53,55,133–135} and knowledge about specific gene products with potential high efficacy for immune priming resulting in disease resistance is needed.

3.2 | Semiochemical production

Another important consideration is the question of what attracts parasites to a particular host or deters parasites from infecting other hosts or non-hosts. Semiochemicals are chemical substances released by one organism that affect the behaviour of other organisms. Semiochemicals that attract another species and harm the emitting host species are known as kairomones whereas those that deter the other species and benefit the emitting host species are known as allomones. Copepodids suspended in the water column detect and show a positive rheotactic response to nearby hosts guided by chemical and mechano-sensory clues.^{136–140} Host-derived kairomones (reviewed in Reference [141]) play a key role in attracting lice at close distances.¹³⁸ Silencing of chemoreceptors in copepodids interferes with recognition of the host.^{142,143} *L. salmonis* is highly host-specific, largely infecting fish of *Salmo* and *Oncorhynchus* genera with different rates of success, whereas *C. elongatus* parasitizes more than 80 marine fish species.¹⁴⁴ This suggests a combination of species-specific kairomones that attract *L. salmonis* to susceptible species (*Salmo* spp.) and allomones that deter *L. salmonis* from the more resistant species including *Oncorhynchus* spp.¹⁴¹ The evidence also suggests that *C. elongatus* attracting kairomones are produced by many species.

Behavioural tests (directional movement of salmon lice and electrophysiology) show that lice are attracted to water conditioned with salmon and that the effect is associated with the volatile fraction.¹⁴⁵ Specific compounds such as isophorone (3,5,5-trimethylcyclohex-2-en-1-one) and sulcatone (6-methyl-5-hepten-2-one) have been identified as candidate lice attracting kairomones for Atlantic salmon.^{140,146}

In addition to variation in attractants in the water among and within species, there is individual variation among hosts with respect to the production of certain compounds in the mucus. Volatile organic compounds including 1-octenol-3-ol, 1-penten-3-ol and sulcatone in the mucus of Atlantic salmon have been found to be associated with increased sessile lice count.¹⁴⁷ These compounds are secondary lipid oxidation products which have been linked to host identification in biting insects such as mosquitoes and midges.^{148–150} The concentration of these compounds in mucus differs between different families of Atlantic salmon (37%–58%) fed the same diet and is associated with higher lice counts in individuals.¹⁵¹

Targeted chemical analysis (e.g.,¹⁵²) of such mucous compounds may allow identification of phenotypes that can be used in selective breeding. More information is, however, needed on the genetic basis and heritability of the production of semiochemicals for this purpose. Using genomic or proteomic data to dissect susceptibility to louse infestation into several underlying and more objective component phenotypes could potentially offer more specific information to complement and improve the accuracy of selection. For instance, semiochemicals that are exuded from the skin in the mucus could be

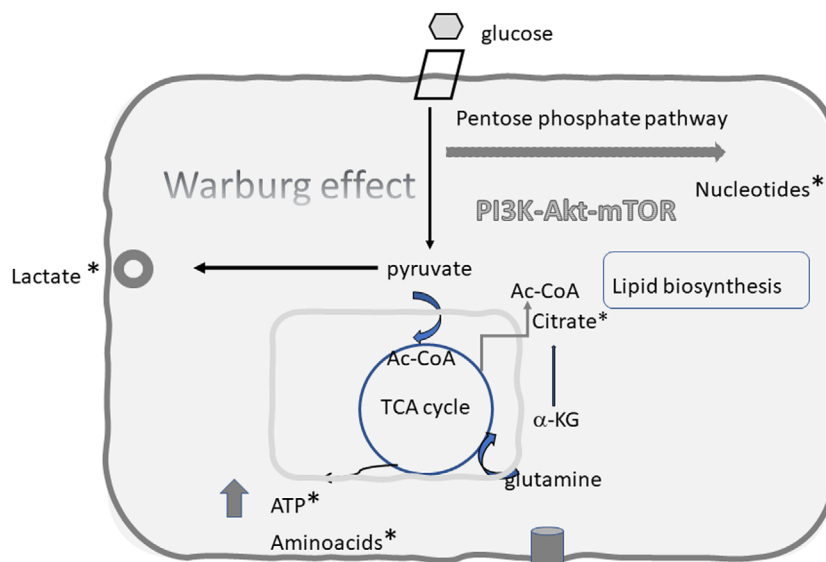


FIGURE 3 Summary of metabolic effects during early (6–12 h) white spot syndrome virus (WSSV) infection. WSSV triggers aerobic glycolysis (Warburg effect) via activation of the PI3K-Akt- mTOR pathway. The hallmark of aerobic glycolysis is the high levels of glucose consumption and lactate production. However other metabolic pathways are also enhanced, including the pentose phosphate pathway (PPP), the lipid metabolism pathway and the glutamine metabolism pathways. These metabolic changes support the high energy requirements of viral replication

sampled non-invasively from the host and could have potential as markers for selection.

improve our understanding of the relationship between each aquaculture species and its pathogens.¹⁷⁷

4 | EXPLORING THE GENETIC FACTORS UNDERLYING HOST RESISTANCE

4.1 | Genomic tool application

Host–parasite and host–viral interactions are extremely complex, and their study requires a holistic approach involving the use of technologies that investigate different aspects of the interaction (Figure 1 and Table 1). Knowledge about the specific genes and mechanisms involved in providing greater disease resistance are needed, particularly for potential gene editing applications downstream. In this section, we provide an overview of the different genomic technologies that can elucidate different components of host–pathogen interactions. While the generation of multiomics datasets is likely to help elucidate host–pathogen interactions and other biological processes, extracting meaningful information from these types of datasets will require appropriate integration methods. The recent explosion of genomics has led to the development of multiple methods for the integration of different data, based on very different approaches ranging from k-means clustering or linear models to complex Bayesian models.¹⁷⁵ The choice of methods to use for the most common integration challenges will depend on the types of data and on the availability of already tailored methods. For instance, single-cell RNA-seq can be easily integrated with single-cell ATAC-seq or spatial transcriptomics using the freely available R package Seurat.¹⁷⁶ The usefulness of these complex datasets will depend on our understanding of gene/protein function. Mapping protein interactions will also provide fundamental information that should help

4.1.1 | Genome-wide association of marker tests

Genome-wide association studies (GWAS) have probably been the most important contributor to the identification of genes affecting disease resistance so far (Tables 1 and 2). These approaches detect specific regions of the genome associated with increased resistance to a disease (QTL), and rely on the existence of genetic variation in resistance among the animals of a population. Resistance is typically measured as morbidity or mortality/survival via controlled disease challenges or natural outbreaks, although more complex phenotypes such as time to death or pathogen load are also frequently used. The final goal is to detect major QTL that can be selected for or against in aquaculture breeding programmes using associated genetic markers (marker-assisted selection). However, QTLs with large effects on the trait of interest are infrequent, and resistance to most diseases is polygenic in nature.²⁴²

Although QTL for ‘lice-attracting salmon’ have been found (www.aquagen.no,⁶⁵), lice resistance measured as lice count and lice density seems to be polygenic.²⁴³ Efforts to dissect the genetic mechanisms behind salmon lice resistance have yielded three chromosomal regions in which there are genes explaining between 7% and 13% of the genetic variation in resistance to *C. rogerresseyi* in Atlantic salmon in Chile.⁶⁵ The effect of individual QTL on resistance is relatively small, and therefore genomic selection is preferred over marker-assisted selection. Knockout or up-regulation of the causative genes via genome engineering (e.g., gene editing) might provide greater resistance than the causative variants themselves. Gene expression data and knowledge from other experiments, some comparing the

TABLE 1 Examples of -omic technologies with potential applications for exploring the genetic basis of host resistance

-Omic technology	Example	Application	Relevance in the study of disease resistance	Key references
Genomics	GWAS	Detection of genomic regions and allelic variants associated with phenotypic variation (e.g., disease resistance)	Fine mapping of loci associated with host resistance providing candidates for further testing (and potentially enabling marker-assisted selection)	[153]
	Gene editing	Study of gene function	Identification and or confirmation of relevant /causative genes involved in disease resistance	[154]
	Genome-wide CRISPR knock-out screens	Assessment of the impact of all the genes in a genome on a phenotype	Unbiased identification of genes involved in disease resistance	[155]
Transcriptomics	RNAseq	Transcriptomic differences between tissue samples	Changes in the transcriptome of a tissue sample in response to infection.	[156,157]
	scRNA-seq	Transcriptomes measured at single-cell resolution	Detects changes in cell populations and gene expression of each cell type in response to infections	[158,159]
	snRNA-seq	Similar to scRNAseq but using nuclei instead of whole cells	As per scRNAseq	[160,161]
	Spatial transcriptomics	Transcripts mapped and quantified to their position in a tissue section	Expression changes at spatial resolution in the tissue section in response to infection	[162,163]
	Perturbation screens	Combination of pooled CRISPR screens and scRNAseq to study the function of multiple genes	Transcriptomic changes induced by the absence or up-regulation of a gene of interest at cellular resolution	[164]
Biomolecule omics	Mass spectrometry	Identifying proteins, carbohydrates, glycans, glycoproteins and metabolites produced by host.	Identify semiochemicals produced by host and parasite-host protein interactions	[165]
	Spatial proteomics	Similar to spatial transcriptomics but for mass spectrometry identification of proteins	How are protein interactions differing in areas of the tissue section responding to infection	[166]
Epigenomics	CHIA-PET or HIC	Generation of genome-wide chromatin interaction maps	Identify genomic regions and sequences showing differential gene regulation	[167,168]
	ATACseq and scATAC-seq	Map chromatin accessibility (potentially at single-cell resolution)	Identify differences in chromatin accessibility are associated with disease resistance.	[169,170]
	CHIPseq	Genome-wide identification of the binding sites of DNA-associated proteins	Identify differences in DNA binding of specific transcription factors and other chromatin-associated proteins associated with disease resistance	[171]
	WGBS	Methylation status at each cytosine base across the genome	Identify differences in DNA methylation associated with disease resistance	[172,173]
	RRBS	As for WGBS but assaying only genomic regions with high CpG content	As for WGBS	[174]

Abbreviations: GWAS, genome-wide association studies; scRNA, Single-cell RNA sequencing; snRNA, Single-nuclei RNA sequencing; CHIA-PET, chromatin interaction analysis with paired-end-tag-sequencing; ATACseq, assay for transposase-accessible chromatin with high throughput sequencing; CHIPseq, chromatin immunoprecipitation sequencing; WGBS, whole-genome bisulphite sequencing; RRBS, reduced representation bisulfite sequencing.

transcriptome of resistant and susceptible Atlantic salmon (e.g.,^{87,90,93}), could be drawn upon to identify candidate genes and potential causative mutations associated with QTL. These could then be edited using CRISPR/Cas9 to generate beneficial variation de novo (see below), potentially with a larger effect than the detected QTL.

GWAS has detected several QTL associated with WSSV resistance (time to death) in *P. monodon*²³⁷ and identified nearby mapping genes that could potentially influence innate immunity. The estimated

size of the effects of these QTL on resistance was relatively small. Some of the single nucleotide polymorphisms (SNPs) mapping to these regions occurred in genes affecting the development and function of the innate immune system such as those in the ubiquitin-proteasome pathway, affecting lymphocyte cell function, heat shock proteins, the TOLL pathway, protein kinase signal transduction pathways, mRNA binding proteins and lectins. Genome scans of broader shrimp populations utilising denser sets of SNP markers mapping to

TABLE 2 Published application of single nucleotide polymorphism (SNP) genome-wide association studies (GWAS) and quantitative trait loci (QTL) scans to explore the genetic basis of host resistance for species in aquaculture for food production

Atlantic salmon
Infectious salmon anaemia ^{178,179}
Pancreas disease ^{180,181}
Piscine myocarditis virus ^{182–184}
Amoebic gill disease ^{185–187}
Sea lice ^{65,188,189}
Infectious pancreatic necrosis ^{81,154,190,191}
<i>Piscirickettsia salmonis</i> ¹⁹²
Coho salmon
<i>Piscirickettsia salmonis</i> ¹⁹³
Common carp
<i>Aeromonas hydrophila</i> ¹⁹⁴
Koi herpesvirus ^{195–197}
Rohu carp
<i>Aeromonas hydrophila</i> ¹⁹⁸
Rainbow trout
<i>Aeromonas salmonicida</i> ¹⁹⁹
<i>Vibrio anguillarum</i> ²⁰⁰
<i>Ichthyophthirius multifiliis</i> ²⁰¹
<i>Piscirickettsia salmonis</i> ²⁰²
Infectious pancreatic necrosis ²⁰³
Columnaris ²⁰⁴
Coldwater bacterial disease ^{205–208}
Infectious pancreatic necrosis ^{203,209}
Viral haemorrhagic septicaemia virus ²¹⁰
Nile tilapia
Tilapia lake virus ²¹¹
Red tilapia
<i>Streptococcus agalactiae</i> ²¹²
European sea bass
Nervous necrosis virus ^{213,214}
Vibriosis ²¹⁵
Asian sea bass
Nervous necrosis virus ^{216,217}
Red spotted grouper
Nervous necrosis virus ²¹⁸
Yellowtail (<i>Seriola quinqueradiata</i>)
Benedenia disease ^{219–221}
Pacu
<i>Aeromonas hydrophila</i> ²²²
Catfish
<i>Aeromonas hydrophila</i> ²²³
Enteric septicemia ^{224–226}
Columnaris disease ²²⁷

TABLE 2 (Continued)

Yellow croaker
<i>Cryptocaryon irritans</i> ²²⁸
<i>Pseudomonas plecoglossicida</i> ²²⁹
Gilthead seabream
Phytobacteriosis ²³⁰
<i>Sparicotyle chrysophrii</i> ²³¹
Japanese flounder
<i>Vibrio anguillarum</i> ²³²
Turbot
<i>Aeromonas salmonicida</i> ²³³
<i>Philasterides dicentrarchi</i> ^{234,235}
Red sea bream
Iridoviral disease ²³⁶
Black tiger shrimp
White spot syndrome virus ²³⁷
Pacific oyster
<i>Vibrio alginolyticus</i> ²³⁸
Ostreid herpesvirus ²³⁹
European flat oyster
<i>Bonamia ostreae</i> ²⁴⁰
Hard clam
Quahog parasite unknown disease ²⁴¹

Note: Disease names or pathogens are listed below each host species (host species in bold). Commonly used names are included.

the QTL regions of interest can be used to verify and narrow the region searched for the occurrence of putative causative genes. Further work is needed to study the response of these and other candidate genes to WSSV infection in susceptible and resistant shrimp and to understand whether the same mechanisms might be involved in other penaeid shrimp such as *L. vannamei*. GWAS has been used in this way as a ‘first pass screen’ to help target future research efforts.

4.1.2 | Single-cell RNA sequencing

Sequence count data, derived from the application of RNA-sequencing (Table 1), has been used to compare the host gene expression response to infection in highly susceptible and resistant individuals for many aquatic diseases of concern to aquaculture (Table 3). In contrast to bulk RNA-seq, single-cell sequencing provides sequences of the transcriptomes of individual cells. Single-cell sequencing has the resolution to identify cell types and transcript changes of interest during a disease challenge that would otherwise be indistinguishable with bulk sequencing⁵⁰² and since being named method of the year by Nature in 2013,¹⁵⁸ single-cell RNA sequencing (scRNAseq) has provided novel insights into infectious disease response and host-pathogen interactions (reviewed in Reference [159]). The most widely used platforms (e.g., 10× Genomics Technology) use microfluidics to capture

TABLE 3 Published application of RNA-seq to explore the genetic basis of host resistance for species in aquaculture for food production

European sea bass
<i>Ceratomyxa oestroides</i> ²⁴⁴
Nodavirus ²⁴⁵⁻²⁴⁷
<i>Vibrio anguillarum</i> ^{248,249}
Japanese sea bass
<i>Vibrio harveyi</i> ²⁵⁰
Largemouth bass
<i>Micropterus salmoides</i> rhabdovirus ²⁵¹
Blunt snout bream
<i>Aeromonas hydrophila</i> ²⁵²
Gilthead sea bream
<i>Sparicotyle chrysophrii</i> ²⁵³
Red sea bream
<i>Vibrio anguillarum</i> ²⁵⁴
Sea bream
Nervous necrosis virus ²⁵⁵
Black carp
<i>Aeromonas hydrophila</i> ²⁵⁶
Catla carp
<i>Edwardsiella tarda</i> ²⁵⁷
Common carp
<i>Aeromonas hydrophila</i> ²⁵⁸
<i>Aphanomyces invadans</i> ²⁵⁹
Cyprinid herpesvirus 3 ^{260,261}
Koi herpesvirus ²⁶²
Spring viraemia of carp virus ²⁶³
Gibel carp
Cyprinid herpesvirus 2 ^{264,265}
Liver myxobolosis ^{266,267}
Grass carp
<i>Aeromonas hydrophila</i> ^{268,269}
Grass carp reovirus ²⁷⁰⁻²⁷⁴
<i>Ichthyophthirius multifiliis</i> ²⁷⁵
<i>Vibrio mimicus</i> ²⁷⁶
Rohu carp
<i>Aphanomyces invadans</i> ²⁷⁷
<i>Aeromonas hydrophila</i> ²⁷⁸
Silver carp
<i>Microcystis aeruginosa</i> ²⁷⁹
African catfish
<i>Aeromonas veronii</i> ²⁸⁰
Channel catfish
Channel catfish virus ²⁸¹
<i>Edwardsiella ictaluri</i> ²⁸²⁻²⁸⁸
<i>Flavobacterium columnare</i> ²⁸⁴⁻²⁹⁵
Enteric septicemia ^{296,297}
<i>Yersinia ruckeri</i> ²⁹⁸

(Continues)

TABLE 3 (Continued)

Ussuri catfish
<i>Aeromonas veronii</i> ²⁹⁹
Yellow catfish
<i>Edwardsiella ictaluri</i> ³⁰⁰
Cobia
<i>Streptococcus dysgalactiae</i> ³⁰¹
Yellow croaker
<i>Pseudomonas plecoglossicida</i> ^{229,302-305}
European eel
<i>Aeromonas hydrophila</i> ³⁰⁶
Rana grylio virus and the <i>Herpesvirus anguillae</i> ³⁰⁷
<i>Anguillicola crassus</i> ³⁰⁸
Japanese eels
<i>Anguillicola crassus</i> ³⁰⁸
Olive flounder
<i>Edwardsiella tarda</i> ^{309,310}
Megalocytivirus ³¹⁰
<i>Hirame novirhabdovirus</i> ^{311,312}
Infectious haematopoietic necrosis virus ³¹²
<i>Listonella anguillarum</i> ³¹³
Lymphocystis disease virus ³¹⁴
Megalocytivirus ³¹⁵
Viral haemorrhagic septicaemia virus ^{312,316-321}
Gibel carp
cyprinid herpesvirus 2 ³²²
Fat greenling
<i>Vibrio harveyi</i> ³²³
Brown-Marbled Grouper
<i>Epinephelus fuscoguttatus</i> ³²⁴
Giant grouper
Spotted knifejaw iridovirus ³²⁵
Malabar grouper
Nervous necrosis virus ³²⁶
Kelp grouper
Nervous necrosis virus ³²⁷
Orange-spotted grouper
Iridoviruses ³²⁸
Nervous necrosis virus ^{329,330}
<i>Pseudomonas plecoglossicida</i> ³³¹⁻³³⁷
Red spotted grouper
Nervous necrosis virus ³³⁸⁻³⁴⁰
Tiger grouper
<i>Spatholobus suberectus</i> , <i>Phellodendron amurense</i> , or <i>Eclipta prostrata</i> ³⁴¹
Yellowtail Kingfish
Gut enteritis ³⁴²
Lumpfish
<i>Vibrio anguillarum</i> ³⁴³

(Continues)

TABLE 3 (Continued)

Mandarin fish
Infectious spleen and kidney necrosis virus ³⁴⁴
Fathead Minnow
<i>Yersinia ruckeri</i> ³⁴⁵
Rare Minnow
Grass carp reovirus ³⁴⁶
Pacu
<i>Aeromonas hydrophila</i> ³⁴⁷
Yellow perch
<i>Apophallus brevis</i> ³⁴⁸
Black rockfish
<i>Photobacterium damsela</i> ³⁴⁹
Atlantic salmon
Infectious salmon anaemia virus ^{178,350–356}
Amoebic gill disease ^{185,357–359}
Salmon and sea lice ^{65,87,351,360–362}
<i>Aeromonas salmonicida</i> ³⁶³
Infectious haematopoietic necrosis virus ³⁶⁴
Infectious pancreatic necrosis virus ^{365,366}
<i>Pilchard orthomyxovirus</i> ³⁶⁷
<i>Piscirickettsia salmonis</i> ^{192,368–370}
Pancreas disease ^{180,371–373}
<i>Saprolegnia parasitica</i> ³⁷⁴
Sockeye salmon
Piscine reovirus and infectious haematopoietic necrosis virus ³⁷⁵
Rhabdovirus ³⁷⁶
Asian seabass
Scale drop disease virus and lates calcarifer Herpes virus ³⁷⁷
Viral nervous necrosis ^{378,379}
Hybrid snakehead
<i>Aeromonas schubertii</i> ³⁸⁰
Snakehead
Vesiculovirus ^{381,382}
Half-smooth tongue sole
<i>Vibrio anguillarum</i> ³⁸³
Senegalese sole
Nervous necrosis ³⁸⁴
Steelhead
<i>Ceratonova shasta</i> ³⁸⁵
Amur Sturgeon
<i>Mycobacterium Marinum</i> ³⁸⁶
<i>Yersinia ruckeri</i> ^{387,388}
Nile tilapia
<i>Saprolegnia parasitica</i> ^{374,389}
<i>Francisella noatunensis</i> ³⁹⁰
Meningoencephalitis ³⁹¹
<i>Streptococcus agalactiae</i> ^{392–401}

TABLE 3 (Continued)

<i>Streptococcus iniae</i> ⁴⁰²
Tilapia lake virus ^{403–405}
Brown trout
Proliferative darkening syndrome ⁴⁰⁶
<i>Tetracapsuloides bryosalmonae</i> ⁴⁰⁷
Rainbow trout
infectious haematopoietic necrosis virus ^{408,409}
<i>Yersinia ruckeri</i> ⁴¹⁰
<i>Aeromonas salmonicida</i> and <i>Ceratomyxa shasta</i> ⁴⁰⁹
<i>Flavobacterium psychrophilum</i> ^{411–413}
Glochidia ⁴¹⁴
<i>Ichthyophthirius multifiliis</i> ^{415–417}
Infectious pancreatic necrosis virus ⁴¹⁸
<i>Piscirickettsia salmonis</i> ^{419,420}
Proliferative kidney disease ⁴²¹
<i>Vibrio anguillarum</i> ⁴²²
<i>Ceratonova shasta</i> ⁴²³
Steelhead trout
<i>Ceratonova shasta</i> ⁴²³
Turbot
<i>Edwardsiella piscicida</i> ⁴²⁴
<i>Enteromyxum scophthalmi</i> ^{425,426}
<i>Vibrio anguillarum</i> ⁴²⁷
Mitten Crab
<i>Micrococcus luteus</i> , <i>Vibrio alginolyticus</i> and <i>Pichia pastoris</i> ⁴²⁸
Mud Crab
<i>Vibrio parahaemolyticus</i> ⁴²⁹
White spot syndrome virus ^{430,431}
Australian red claw crayfish
<i>Aeromonas veronii</i> ⁴³²
Red swamp crayfish
Infectious hypodermal and haematopoietic necrosis virus ⁴³³
<i>Aeromonas hydrophila</i> ⁴³⁴
<i>Vibrio cholerae</i> ⁴³⁵
WSSV and <i>Aeromonas hydrophila</i> ⁴³⁶
Far eastern mussel
<i>Vibrio alginolyticus</i> ⁴³⁷
Bannana shrimp
Hepatopancreatic parvo-like virus ⁴³⁸
White spot syndrome virus ⁴³⁹
Black tiger shrimp
Acute hepatopancreatic necrosis disease ^{440,441}
Decapod iridescent virus 1 ⁴⁴²
Hepatopancreatic necrosis disease ⁴⁴³
White spot syndrome virus ⁴⁴⁴
Chinese grass shrimp
<i>Tachaea chinensis</i> ⁴⁴⁵

TABLE 3 (Continued)

Chinese mitten crab
White hepatopancreas syndrome ⁴⁴⁶
Fairy Shrimp
Bacterial black disease ⁴⁴⁷
Giant fresh water prawn
<i>Enterobacter cloacae</i> ⁴⁴⁸
Nodavirus ⁴⁴⁹
<i>Vibrio parahaemolyticus</i> ⁴⁵⁰
White spot syndrome virus ⁴⁵¹
Kuruma shrimp
<i>Vibrio alginolyticus</i> ⁴⁵²
White spot syndrome virus ^{453–455}
Whiteleg shrimp
<i>Spiroplasma eriocheiris</i> ⁴⁵⁶
Taura syndrome virus ^{457,458}
<i>Vibrio parahaemolyticus</i> ^{459–465}
White spot syndrome virus ^{466–474}
Manila clam
<i>Perkinsus olseni</i> ^{475,476}
<i>Vibrio anguillarum</i> ⁴⁷⁷
<i>Vibrio tapetis</i> ⁴⁷⁸
Brown muscle disease ⁴⁷⁹
Surf clams
<i>Vibrio</i> spp. ⁴⁸⁰
Pacific oyster
<i>Vibrio splendidus</i> ^{481,482}
Norovirus ⁴⁸³
Ostreid herpesvirus ^{135,484,485}
<i>Staphylococcus aureus</i> ⁴⁸²
Pearl oyster
<i>Vibrio alginolyticus</i> ⁴⁸⁶
Blackfoot paua
Abalone viral ganglioneuritis ⁴⁸⁷
Coloured abalone
<i>Haliotid herpesvirus-1</i> ⁴⁸⁸
Malacoherpesviruses ⁴⁸⁹
Chinese razor clam
<i>Vibrio parahaemolyticus</i> ^{490,491}
Spotted hard clam
<i>Vibrio parahaemolyticus</i> ⁴⁹²
Cupped oyster
Ostreid herpesvirus 1 ⁴⁹³
Eastern oyster
<i>Perkinsus marinus</i> ^{494–496}
American oysters
<i>Perkinsus marinus</i> ⁴⁹⁷
Roseovarius oyster disease ⁴⁹⁸

(Continues)

TABLE 3 (Continued)

Japanese sea cucumber
<i>Vibrio splendidus</i> ⁴⁹⁹
Sea urchin
Spotting disease ⁵⁰⁰
<i>Vibrio</i> sp. ⁵⁰¹

Note: Disease names or pathogens are listed below each host species (host species in bold). Commonly used names are included.

individual cells with uniquely barcoded beads, each of which are covered with unique molecular identifiers that tag each individual RNA molecule within a cell. This combination of barcodes allows tracing of each RNA molecule to its cell of origin.¹⁶⁰

This technique can also be applied on single nuclei (snRNAseq) instead of whole cells. The main downside of this being the loss of cytoplasmic RNA⁵⁰³ which results in lower numbers of genes per cell sequenced, and possible subsequent loss of well-known marker genes. However, snRNAseq has the advantage of allowing samples to be flash frozen for storage, transportation, nuclei extraction and sequencing at a later date. Unlike whole cells, nuclei dissociation can be performed on ice, minimising heat stress and the subsequent loss of specific cell types.^{160,161,504} Robust nuclei dissociation protocols have been developed and shown to work well in different tissue types and produce results concordant with the sequencing of whole cells.^{505,506}

Single-cell/nuclei RNA sequencing can identify changes in cell type, abundance, and transcripts between samples. By identifying transcriptomic signatures at the cellular level these technologies should provide a much more precise molecular understanding of disease resistance in aquaculture species than has been previously possible. For instance, single-cell RNA sequencing has been used to characterise the immune system of the shrimp *Marsupenaeus japonicus*, identifying six different types of haemocytes with differentiated immune roles.⁵⁰⁷ Only a handful of other single-cell studies have been carried out in aquaculture species, for example, on the immunologic profile of Atlantic salmon gill,⁵⁰⁸ leukocyte populations in *Nile tilapia*⁵⁰⁹ and cold tolerance signatures in the hepatopancreas of whiteleg shrimp.⁵¹⁰ Single-cell technologies therefore have great potential for investigating the biological mechanisms underlying the phenotypic differences in disease response between species.

4.1.3 | Spatial transcriptomics

Spatial transcriptomics is an overarching term for methodologies assigning mRNA to their relative position in a tissue section (Table 1,^{162,163}). These technologies are primarily categorised as either imaging-based approaches (in situ sequencing and in situ hybridization-based methods), or next-generation sequencing (NGS)-based methods. While imaging-based spatial transcriptomic techniques are typically limited to detecting a handful of genes that have been selected a priori, NGS methods indiscriminately target RNA within a spatial area which are mapped back to their spatial position using unique barcodes.^{511–515} NGS methods are therefore an attractive

approach for explorative research, allowing for the spatial depiction of all gene activity in a tissue sample. In terms of ectoparasites such as sea lice, NGS methods could be used to compare fine-scale localised gene expression at the site of parasite attachment. Applying the technique on vertical skin sections through the attachment site of the parasite may capture different responses between resistant and susceptible animals throughout the skin layers. For instance, immune cell infiltration is a well-characterised feature of coho salmon resistance to sea lice,^{72,104,516} but we do not know what signals drive this response in coho, and why this response is weaker in susceptible salmon species. Spatial transcriptomics could be used to discriminate what signals are released by host cells at the attachment site at critical times post-attachment, some of which might be key signals driving immune cell infiltration. Given that the spatial resolution of NGS methods is often greater than the diameter of a single cell, integrating spatial transcriptomic data with snRNAseq/scRNAseq data could facilitate powerful insights into transcriptomic differences by cell type through space.⁵¹⁵ Tissue and cell level spatial proteomics is also possible⁵¹⁷ and could be compared with spatial transcriptomic data to investigate connections between the transcriptome and proteome with respect to disease resistance.

4.1.4 | Proteomics

The study of infectious diseases requires defining the cellular and molecular mechanisms at the site of pathogen entry or attachment to a host. The interacting proteins of both species are likely to have a crucial role in the early infection process. Proteomics can decipher host-pathogen cross-talk, providing vital information on infection, early pathogenesis, host-immune response and mechanisms by which pathogens evade host defence. Use of proteomics in aquatic species disease research is outlined in a review by Moreira et al.⁵¹⁸ Recent years have seen major advancements in the technology, although most new developments have so far been applied to unravelling infectious disease mechanisms and pathogenesis in non-aquaculture species, such as humans. Here, we highlight some of the advanced proteomic tools that will likely have application for investigating infectious disease biology in fish and shellfish.

Proteomics provides a vast array of tools, not only to measure the abundance levels of thousands of individual proteins across various states/conditions from nanogram levels of samples, but also for addressing challenges at proteoform and post-translational modification levels. Global quantitative profiling employing stable isotopic labelling (e.g.,⁵¹⁹) and extensive fractionation of peptides prior to mass spectrometry (MS) by various chromatography techniques give deep proteome coverage.

New generation high-resolution MS systems combining additional gas-phase separation of peptide ions based on ion-mobility and therefore label-free quantification approaches by either data-dependant or data-independent acquisition methods⁵²⁰ can be used to quantify proteins over a larger dynamic range from complex mixtures. Targeted MS methods can be used to study a set of proteins of interest in complex mixtures with high sensitivity, selectivity and quantitative reproducibility (e.g., with parallel-reaction monitoring⁵²¹).

Post-translational modifications (PTM) often play a critical role in the biology of infection. It has been widely believed that only eukaryotes were able to make PTMs, however, now high-resolution mass spectrometry has identified a plethora of PTMs in prokaryotic pathogens.⁵²² While host cells can utilise a cascade of PTM changes to accelerate immune response, infecting virus also use PTMs to breakthrough host defence.⁵²³ UNIMOD, a database for PTMs detected by mass spectrometry has now reported over 1500 modifications, indicating the diversity and richness of post-translational regulation, although the biological functions of many of these modifications are unknown (https://www.unimod.org/modifications_list.php, 30 May 2022).

Interactions between host and pathogen proteins are critical for pathogen replication, and for escape from and control of the host immune response. High-resolution LC-MS-based proteomics (e.g., using co-immunoprecipitation with target-specific antibodies conjugated to protein A/G affinity beads followed by MS-based identification,⁵²⁴ proximity-dependent biotin identification^{525,526} with GFP binding nanobodies,⁵²⁷ cofractionation MS,⁵²⁸ clear-native PAGE⁵²⁹ and/or cross-linking mass spectrometry⁵³⁰) generate detailed quantitative protein profiles that could be used to reveal potential immunomodulatory host-parasite interactions.

In *P. vannamei*, GST-pull down and mass spectrometry analysis were used to investigate the interaction of the immediate-early WSSV protein (IE-1) with shrimp proteins, finding 361 host proteins that could potentially bind to IE.⁵³¹ Most of these proteins were involved in signalling pathways such as the prophenoloxidase (proPO), PI3K-AKT, MAPK, focal adhesion, and cell cycle systems. Knockdown of IE-1 reduced viral load and WSSV gene expression, while recombinant IE-1 inhibited host prophenoloxidase in a dose-dependent manner.⁵³¹ Metabolic studies of gill, haemolymph and hepatopancreas have revealed clearly different profiles in WSSV infected and non-infected shrimp, consistent with changes in osmoregulation in the gills, upregulation of the glutathione pathway, increased production of an antimicrobial peptide itaconic acid in the hemolymph and a shift from aerobic to anaerobic metabolism as previously described.⁵³² Interestingly, some of the changes in the haemolymph (increased TCA intermediates and decreased amino acids) were similar to those in haemolymph of *P. vannamei* challenged with *Vibrio parahaemolyticus*⁵³³ while some molecules were specific for each pathogen such as itaconic acid in WSSV-challenged shrimp and increased phosphoenolpyruvic acid (PEP) in shrimp exposed to *V. parahaemolyticus*, suggesting that there are pathogen-specific innate immune responses in shrimp. The use of host-targeted drugs that act on these pathways hijacked by viruses is now emerging as a potential source of antiviral treatments for human medicine.⁵³⁴

4.2 | Semiochemicals

4.2.1 | Detection and characterisation

In marine ecosystems, obligate ectoparasites, such as sea lice, use chemical cues and other sensory signals to increase the probability of encountering a host and to identify appropriate hosts on which they

depend to complete their life cycle. The chemical compounds that underlie host identification by the sea lice are not fully described or characterised. Identification of semiochemicals attracting parasites opens new possibilities for parasite control. Quantitative assays of parasite attractants (kairomones) or repellents (allomones) could guide the selection of favourable variants for breeding programs. The effect of rearing conditions and feeds on attractiveness can be mechanistically assessed. Moreover, semiochemicals can be used in push-pull strategies¹⁴¹ using anti-parasite fouling devices, ointments and baited traps.⁵³⁵ Traditionally semiochemicals are identified by bioassay-guided fractionation. Crude extracts are separated into finer fractions based on chemical properties, for example, polarity, size, or volatility. Active fractions (proven by bioassay) are further separated into new fractions and the procedure is repeated until an active compound has been isolated. High-resolution analytical methods are often now used in combination with statistical methods (metabolomics) to identify and directly bioassay candidate compounds.^{535,536} The metabolomic approach can be used to identify candidate compounds involved in host-parasite interactions, and to distinguish those produced exclusively, or at markedly higher levels, by susceptible species or individuals. The effect of candidate compounds on the behaviour of parasites at infectious stages of development can then be assayed. To accentuate differences and improve the power of detection, samples from host individuals with extremely high and low levels of resistance to the parasite (based on estimated breeding values and/or direct counts post-infection) could be tested. Non-target species that are not parasitized can be used as negative controls. Candidate compounds may be water soluble metabolites released from the fish directly into seawater¹⁴⁵ or compounds secreted in the mucous or released as faeces or urine. A full spectrum of semiochemicals could be obtained from water bathing the host using solid phase extraction.¹⁴⁵ Sub-nanogram quantities of the semiochemicals in the collected samples can be measured using a gas chromatography-flame ionisation detector or -mass spectrometer multiple-point external method with authentic standards of identified compounds.

Replicate filming tanks or arenas populated with copepodids have been used for rapid screening of behavioural responses (swimming speed, hop frequency, and rate of change of direction in response) to measure lice attraction/repulsion to compounds.⁵³⁷ The responses of lice to compounds in the behavioural tests are corroborated by neurophysiology experiments.¹³⁹ Copepodids are stimulated to swim rapidly upward using a standardised light flash (generating a change of light intensity simulating a group of fish swimming overhead). Compounds of interest are added to the water at higher than natural concentrations to determine if they enhance the response to the standardised light stimulus. The behavioural data is digitally recorded and evaluated using motion analysis equipment that has been specifically developed for measuring such lice behavioural differences.^{143,146,537,538} Many different compounds can be processed relatively quickly using this assay. Different concentrations of the validated lice-attractant compounds can be used to generate stimulus-response curves. These stimulus-response curves serve as a basis to estimate the distance from a point source of salmon at which the compounds would activate free-swimming copepodids.

Another line of research involves testing if exposure to semiochemicals provokes differential gene expression in copepodids (e.g., induces the production of immunomodulatory compounds). Salmon conditioned water and putative semiochemicals could be tested on batches of infective copepodid larvae. Exposed copepodids could be sampled at specific time points post-exposure and their transcriptomic profile analysed using Illumina bulk RNASeq. Profiling of treated and untreated lice would inform as to whether the levels of louse host-interacting proteins identified are also influenced by semiochemicals released by resistant or susceptible salmonid hosts.

To associate semiochemicals affecting host-lice interactions with candidate genes affecting semiochemical production, a focus could be on genes coding for enzymes involved in pathways that produce secondary metabolites. These may be absent/present in susceptible salmon species, differentially regulated or absent/present in resistant salmon species and absent/present in other marine species. In common with all lipophilic compounds, the metabolism of semiochemicals most likely involves cytochrome P450 (CYP) monooxygenases. The CYP superfamily is characterised by extremely rapid evolution and diversification, and frequent acquisition and loss of genes.^{539,540} Some CYPs (e.g., the entire CYP2M1 subfamily) are found only in salmonid fish.⁵⁴¹ Tissue profiles combined with gene expression data from existing transcriptome databases (e.g.⁵⁴²) and qPCR could be used to identify target genes for blocking the production of parasite attractants.

4.3 | Gene editing to identify functional disease resistance genes

4.3.1 | Targeted editing of candidate loci in embryos and cells

The CRISPR-Cas system⁸⁵ is a powerful and versatile genome editing tool that can be used to knock out, knock in or to modify transcriptional regulation of target genes. Knock out of targets can be achieved by delivering a Cas enzyme such as Cas9 or Cas12a together with a guide RNA (gRNA) which guides the Cas enzymes to bind the target region and make double-strand breaks (DSBs), which are typically repaired via non-homologous end joining resulting in random insertion or deletion mutations (indels) around the DSB sites.⁵⁴³ Knock in, or other forms of precise editing, can be achieved by adding template DNA of the desired sequence containing homology arms together with the Cas and gRNAs, such that the DSBs are followed by homology-directed repair (HDR).⁵⁴³ The transcriptional regulation of targets can be achieved by using modified dead Cas9 (dCas9) with transcriptional activators or suppressors for upregulation or suppression of the expression of targets.⁵⁴⁴ Finding an efficient method for the delivery of editing constructs into cells or embryos is critical for successful gene editing. There are three general pathways by which the components for gene editing can be delivered: (1) physical administration by microinjection, electroporation or hydrodynamics, (2) viral vector delivery or, (3) non-viral vector delivery using, for example, liposomes.⁵⁴⁵ In addition, editing efficiency varies

depending on gRNA sequence and is also influenced by features associated with the target sequence such as chromatin state which affects accessibility to the target region.⁵⁴⁶ So far there have been few applications of gene editing to species in aquaculture (Table 4). CRISPR-Cas9 has been successfully used for both the knock out and in of genes in Atlantic salmon in vivo by microinjection into the zygote, and development of edited animals is becoming relatively routine^{556–560} as are knockouts in salmonid cell lines using electroporation of Cas ribonucleoprotein or using lentiviral delivery.^{561,562} In contrast, attempts to perform CRISPR-Cas9 genome editing in *L. vannamei* shrimp zygotes were unsuccessful using electroporation and chemical transfection.⁵⁶³ Further research on delivery methods of CRISPR-Cas is required to achieve efficient in vivo genome editing in penaeid shrimp, for example via microinjection of CRISPR-Cas9, as has been successfully applied to the decapod (non-penaeid) shrimp *Exopalaemon carinicauda*.^{555,563–568}

Based on results from large-scale and integrated genomic analyses, target genes could be chosen for the investigation of their functional role using genome editing tools. These targets could be candidate genes or pathways purported to underlie intra- or inter-specific genetic variation in host resistance to the pathogen, for example, candidate genes within a QTL region. The targets may also be identified from knowledge of the biology of the host–pathogen interaction, for example, as was the case that led to the knockout of a domain of a cellular receptor causing complete resistance to the viral disease porcine reproductive and respiratory syndrome in pigs.⁵⁶⁹ Target loci for host resistance to sea lice might be those (i) upstream of immune pathways involved in successful lice rejection, (ii) putative targets of louse immunomodulation, or (iii) enzyme(s) required for the production of semiochemical(s) that are found to differ (in occurrence or gene expression) between salmon species with differing louse attachment or activity. Similarly, targets for WSSV host resistance would include (i) genes involved in the adhesion and entrance of WSSV into host cells such as the newly cloned chondroitin proteoglycan 2 of *Litopenaeus vannamei* (LvCPG2), which interacts with both VP26 and VP28 of WSSV facilitating WSSV adhesion and penetration into shrimp hemocytes,⁵⁷⁰ (ii) the family of NAD⁺-dependent protein deacetylases sirtuins, that can regulate viral replication in vertebrates (in *P. vannamei* the silencing of LvSIRT1 was associated with a decreased gene expression in WSSV⁵⁷¹) or, (iii) genes involved in activation of Toll, IMD signalling and JAK/STAT pathways, (iv) other genes affecting the up-regulation or effectiveness of AMPs, (v) enhancers of NF-κB signalling and, (vi) inhibitors of the PI3K-Akt- mTOR pathway. Gene knock out using CRISPR-Cas9 targeting early exons, or gene upregulation via dCas9 with a transcriptional activator, could be used as appropriate.

High throughput genome-wide CRISPR screens may also be an effective route to use for identifying targets (Table 1). CRISPR-Cas mediated genome-wide gene functional screenings such as genome-wide CRISPR knockout (GeCKO), CRISPR activation (CRISPRa) or interference (CRISPRi) screenings could also be used to identify genes involved in disease resistance.⁵⁷² To perform such screenings, engineered cell lines expressing appropriate Cas effectors, and a valid way to deliver gRNA libraries into the cells, are prerequisites. To date, lentiviral delivery has been optimised in salmonid cell lines⁵⁶¹ but

TABLE 4 Application of gene editing for boosting host resistance for species in aquaculture for food production. Commonly used names are included

Host species	Pathogens	Target editing
Atlantic salmon	Infectious pancreatic necrosis virus ¹⁵⁴	Knockout of NEDD-8 activating enzyme 1 (<i>nae1</i>) and epithelial cadherin (<i>cdh1</i>)
Asian seabass	Nervous necrosis virus ³⁷⁸	Knockout of ribonucleoside-diphosphate M1 (<i>rrm1</i>)
Channel catfish	<i>Edwardsiella ictaluri</i> ^{547,548} <i>Flavobacterium columnare</i> ^{547,548}	Transgenesis of <i>cecropin</i> , knockout of toll/interleukin 1 receptor domain-containing adapter molecule (<i>ticam1</i>) and rhamnose binding lectin (<i>rb1</i>)
	Bacterial (e.g., <i>Acinetobacter baumannii</i> and <i>Klebsiella pneumoniae</i>) ⁵⁴⁹	Knock-in of alligator cathelicidin
Grass carp	Grass carp reovirus ^{550,551}	Knockout of junctional adhesion molecule-A (<i>gcJAM-A</i>)
	<i>Aeromonas hydrophila</i> ⁵⁵²	Transgenesis of human lactoferrin (hLF)
Rohu	Viral, bacterial, lice ⁵⁵³	Knockout of toll-like receptor 22 (<i>trl22</i>)
Rainbow Trout	<i>Aeromonas salmonicida</i> and infectious haematopoietic necrosis virus ⁵⁵⁴	Transgenesis of <i>cecropin</i> P1 and synthetic <i>cecropin</i> B analogue (CF-17)
	<i>Aeromonas salmonicida</i> , Infectious Haematopoietic Necrosis Virus and <i>Ceratomyxa shasta</i> ⁴⁰⁹	Transgenesis of <i>cecropin</i> P1
Ridgetail white prawn	<i>Vibrio parahaemolyticus</i> and <i>Aeromonas hydrophila</i> ⁵⁵⁵	Knockout of chitinase

genome-wide CRISPR screenings have not yet been reported in aquaculture species.

4.3.2 | Testing the effects of edits on host resistance

Genome-edited hosts, along with unedited and mock-edited control animals of the same families can be challenged with the pathogen of interest to assess whether the gene(s) subjected to editing affect host

resistance by measuring and comparing host survival and/or pathogen load, as has been done to explore the effect of the chitinase gene on ridge back white shrimp host resistance to *Aeromonas hydrophila* and *Vibrio parahaemolyticus*.⁵⁵⁵ DNA and RNA samples would be collected to check if the edit is present in the target organ and if it has the desired effect on gene expression. Tests for mosaicism (using PCR and sequencing) and checks for obvious effects on the external phenotype and behaviour should also be carried out. Detected signs of developmental abnormalities or high embryonic mortality rates will also inform whether particular edits should be pursued further. A full evaluation of whether there are side effects on other important traits (e.g., off-target effects caused by unintended editing at other sites in the genome or ill-effects resulting from disruption of the target gene) would require growing edited animals until harvest size.

5 | TECHNOLOGIES AND APPROACHES FOR BOOSTING HOST RESISTANCE

5.1 | Genomic selection

Genomic selection,⁸³ which has been described by Goddard and Hayes⁵⁷³ as a form of marker-assisted selection in which genetic markers covering the whole genome are used so that all QTL are in linkage disequilibrium with at least one marker, is now routinely applied to some aquaculture breeding programmes (Table 5).

For traits measured on sibs or other relatives of the breeding candidates, that is, not on candidates themselves, genomic selection is expected to outperform traditional selection methods because it enables selection within families (resulting in better utilisation of genetic variance and a higher intensity of selection) and is more accurate for estimating breeding values. This is especially important for aquaculture species with a large number of fullsib candidates per family⁵⁸⁸ and increases the rate of genetic gain that is possible for a given rate of inbreeding. To use genomic selection instead of traditional pedigree-based breeding value estimation has been shown to increase accuracy in aquaculture breeding schemes.⁵⁸⁹ Genotyping costs for genomic selection can be expensive due to the genotyping of the many individuals in the reference population (to achieve high accuracy), and large numbers of candidates (to achieve a high selection intensity). Therefore, approaches to reduce genotyping costs without compromising prediction accuracy are needed. Designs that aim to reduce genotyping by pooling extreme groups of individuals, such as those in which sibling DNA samples are pooled according to challenge test results, and those utilising genotype imputation, have been found to be effective.^{590,591} Finally, the effects of different marker densities have been tested and optimised (e.g.,⁵⁹²). Estimation of breeding values for the selected candidates are based on the SNP effects estimated in siblings using GBLUP,⁵⁹³ various Bayesian methods or SNP BLUP.⁸³ Single-step genomic BLUP methods that combine information from genotyped and non-genotyped individuals are also used.⁵⁹⁴ The integration of QTL information with genomic

TABLE 5 Application of genomic prediction and/or genomic selection for boosting host resistance for species in aquaculture for food production

Atlantic salmon	Rainbow trout
Amoebic gill disease ^{186,187}	Bacterial cold-water disease ^{574,575}
Sea lice ^{66,576}	<i>Piscirickettsia salmonis</i> ⁵⁷⁷
Coho salmon	Infectious pancreatic necrosis ⁵⁷⁸
<i>Piscirickettsia salmonis</i> ¹⁹³	Haematopoietic necrosis virus ⁵⁷⁹
European sea bass	Red tilapia
Nervous necrosis virus ^{214,580,581}	<i>Streptococcus agalactiae</i> ²¹²
<i>Vibrio anguillarum</i> ⁵⁸²	Striped catfish
<i>Vibrio harveyi</i> ⁵⁸⁰	<i>Edwardsiella ictaluri</i> ⁵⁸³
Gilthead sea bream	Tiger pufferfish
Nervous necrosis virus ⁵⁸⁰	Heterobothriosis ⁵⁸⁴
Phytobacteriosis ²³⁰	Whiteleg shrimp
Pasteurellosis ⁵⁸¹	White spot syndrome virus ⁵⁸⁵
<i>Sparicotyle chrysothrix</i> ²³¹	Pacific oyster
Japanese flounder	Ostreid herpesvirus ⁵⁸⁶
Edwardsiellosis ⁵⁸⁷	

Note: Disease names or pathogens are listed below each host species (host species in bold). Commonly used names are included.

selection, and possible implications on the accuracy of predicted breeding values is a scenario under investigation.¹⁸⁵

Choice of genotyping method can also be used to maximise the cost-benefit ratio. SNP chips are often used. These have some development costs, but give high SNP density, repeatability and reliability. For small populations, where the investment to develop a SNP chip may be high compared to the economic benefits of genomic selection, lower-cost genotyping methods using lower numbers of SNPs have been shown in simulations to give high accuracy of EBV estimation when combining SNP and pedigree information⁵⁸⁸ or when using imputation to higher density SNP panels⁵⁹⁵ (depending on that some animals in the population have been genotyped for higher density). Genotyping-by-sequencing approaches such as RADseq can be used for species where no reference genome exists and/or if SNPs have not previously been detected.⁵⁹⁶ The genotyping needed for genomic selection may also provide practical benefits since young animals can be pooled at an early stage and relationships can be inferred later after genotyping, reducing the need for keeping families separate and for physical tagging.

Genomic selection has provided a powerful and accurate means of achieving high rates of genetic gain for WSSV resistance in *L. vannamei* shrimp.⁵⁸⁵ To run a traditional sib-selection for WSSV resistance, families would have to be kept separate and then tagged before running the challenge test. Keeping families separate would introduce a common environmental effect of family confounded with genetics and is practically challenging requiring more facilities, space

and physical tagging of young shrimp before the WSSV test could be performed. In this case, genomic selection not only increases accuracy and genetic gain, but it opens a possibility to select for traits that would not be feasible to include in a traditional breeding programme. For the case study of WSSV resistance in *L. vannamei*,⁵⁸⁵ one generation of genomic selection increased WSSV survival by 13 percentage units.

5.2 | Gene editing

Application of gene editing in aquaculture breeding requires comprehensive consideration and discussion from technical, regulatory and public acceptance standpoints. Therefore, identification of a strong candidate to edit can be considered as just the first step towards application. For disease resistance traits, various genomic techniques can be applied to identify strong candidate genes, and their function can be investigated in vitro and in vivo using CRISPR-Cas genome editing systems (as described above). If a particularly promising candidate edit is identified, gene editing may then also be applicable as a tool for the incorporation of favourable edits into aquaculture stocks, potentially resulting in a large genetic gain.⁵⁹⁷ Target edits could include de novo alleles identified in closely related species and need not be limited to naturally occurring polymorphisms segregating in commercial populations (e.g., causative mutations for major QTLs).^{597,598} For example, genome editing could potentially enable transfer of the mechanisms of host resistance to sea lice from coho to Atlantic salmon, via modification of specific genes and pathways in Atlantic salmon to mimic the resistance mechanisms found in coho.⁵⁹⁸ The use of gene editing to create de novo alleles could potentially improve the resistance of populations beyond what could be achieved in a short time horizon via selective breeding alone.

For species with relatively long generation intervals (such as Atlantic salmon, 3–4 years), genome editing could be particularly beneficial for speeding up the development of disease resistance. However, because there are normally many traits of importance to aquaculture, and because inbreeding and loss of genetic diversity can reduce the fitness of populations, and both therefore need to be carefully limited, the propagation and dissemination of genome-edited fish will almost certainly need to take place as part of a well-managed selective breeding programme (rather than replacing such a programme). As such, the practicalities of how to incorporate editing technologies into modern aquaculture breeding programmes require careful thought and study. For example, mosaicism remains a major issue with the direct editing of animals via microinjection, and therefore obtaining germplasm fixed for a single desirable edit is both challenging and time consuming. Furthermore, it is conceivable that genome-edited animals may be required to be sterilised to avoid any risk of interbreeding with wild conspecifics. Sterility can also be achieved via genome editing, but it is a particularly challenging trait to include into a breeding programme for obvious reasons, unless it can be easily reversed when reproduction is required. The use of germ cell technologies and surrogate broodstock may offer some potential

solutions, via culture and editing of germ cells in vitro followed by gamete production from surrogate hosts.⁵⁹⁹ This approach could also disentangle the production and dissemination of edited germplasm from the breeding nucleus germplasm, such that sterility could be introduced, and edits could be tailored according to the producers' requirements. Finally, a major challenge after finding targets for editing is to determine whether the editing of such target genes otherwise impacts the biology of the animal. As an initial screen for off-target effects, CRISPR-Cas9 mediated gene functional analysis could be performed first in cell lines.⁵⁶² In any case, further testing and optimisation of methods for accurate and efficient implementation of genome editing for aquaculture will be required before edited fish or shellfish can be widely disseminated onto farms and before any edited animals can be accepted and approved for human consumption.

5.3 | Semiochemicals

5.3.1 | Feasibility as a phenotype for selective breeding

If semiochemical profiles are sufficiently correlated with lice density, then these profiles may have potential as a convenient phenotype for assessing lice susceptibility on breeding candidates and their sibs without challenge testing using lice. Targeted assay(s) for chromatographic analysis of semiochemicals from mucus samples could be developed for compounds that elicit proven behavioural responses in lice. Use of such assays might potentially enable avoidance of the need for costly challenge tests and large field trials. Tandem mass spectrometry using LC-QQQ-MS provides a suitable instrumental platform for this type of measurement. Intraclass correlation coefficients (i.e., the repeatability) of the repeated semiochemical profiles prior to the lice infection could be used as a measure of the temporal consistency of the semiochemical profiles released by the fish over time. The magnitude of the genetic correlation of lice density with the pre-infection semiochemical profiles, and heritability estimates for each semiochemical, will determine whether some of these semiochemicals may be suitable phenotypic measures for indirect selection for increased lice resistance, or for use in other lice-combatting strategies.

5.3.2 | Application of synthesised or extracted semiochemicals

Cost-effective semiochemical production opens up new methods of reducing host–parasite encounters. Some existing commercial feeds are claimed to contain repellent semiochemical additives and European patents exist for some methods and feed compositions for masking fish semiochemicals (e.g., European Patent Number EP2517568A1). Research has shown that ectoparasite infections on fish (amberjack, Nile tilapia and rainbow trout) are reduced when the fish are grown using diets containing plant derived extracts or beta-

glucan additives^{600–603} and proof-of-concept evidence has shown that botanically-derived materials can be incorporated into feed additives to reduce the attraction of *L. salmonis* to salmonid host cues.⁶⁰⁴ Increased knowledge about genetic differences in semiochemical production between resistant and susceptible hosts to sea lice infection may help in the design of the formulation of feed additives for use to boost Atlantic salmon's ability to resist sea lice infection.

Several strategies could be used to design feed additives with potential to boost host resistance to sea lice. For example, ingredients that block odour receptors in the parasite, mask kairomones released from the host, and/or increase allomone production in the host could be tested. As a first pass test, ex vivo fin assays could be used to assess the anti-attachment properties of a candidate dietary ingredient.⁶⁰⁵ Briefly, infectious copepodids are incubated with a candidate ingredient added to the water at different doses together with excised pectoral fins of Atlantic salmon. Copepodids readily attach to submerged fins under these conditions while the test ingredient may reduce the attachment rate if it possesses anti-lice properties. The copepodids' phenotype (number successfully attached to submerged ex-vivo fins, free swimming and moribund animals), as well as copepodid gene expression, could be used to assess the impact of several additives in a mid-throughput manner and help in the prioritisation of candidate ingredients for further lice challenge trials. A more time and resource-demanding variant of the ex vivo fin assay involves a short feeding in vivo trial that precedes the ex vivo fin assay. Atlantic salmon are exposed to the feed that contains the ingredient with suspected anti-attachment properties, or the control feed, for 7–14 days prior to the ex vivo experiment to allow for the bioactive component (s) contained in the anti-lice ingredient to accumulate in skin, including fins, where it acts as a repellent. Pectoral fins from the anti-attachment feed group and control dietary group are removed from fish and placed in pairs (test and control) in glass containers filled with sea water and incubated with *L. salmonis* copepodids. Lower number of lice settling on fins originating from fish exposed to the test feed suggests that the tested dietary ingredient changes olfactory (or nutritional) properties of skin and mucus thus negatively affecting the early phases of lice attachment.⁶⁰⁶ Fin assays allow investigations of early processes that occur in copepodids around the time of attachment in a time-series with many multiple time points.

Full trials to test the effect of feed additives are typically around 3-month long, as they involve host acclimation, pre-feeding of test diets (depending on the rate of accumulation of bioactives in skin and mucus, this period may need to last for up to a month) and an infection challenge with *L. salmonis* copepodids. Different dietary doses in triplicate tanks would need to be tested against the control dietary group. Growth rate and other important traits would be monitored throughout the trial to allow for the detection and assessment of any possible negative effects caused by the tested ingredient. All treated fish would be challenged with lice and resulting infestation densities recorded and compared to control groups to allow for the utility of feed additives for repelling lice and masking salmon to be assessed.

An alternative to boosting host resistance through anti-attachment feeds is to deploy semiochemicals beyond the host to reduce

the number of lice entering sea cages. This might be achieved using baiting traps around the perimeter of the farm, slow-releasing decoy semiochemicals at a distance from the farm, or slow-releasing repellent/masking semiochemicals within the farm footprint to discourage lice. Behavioural responses to host/non-host cues are well-documented in the laboratory,^{136,140} yet the best evidence for efficacy in the sea was produced by a trial of cages with semiochemical-impregnated mesh⁶⁰⁷ reviewed in Reference [46]. Applications of semiochemicals outside of the host may not work at commercial scale, simply because the swimming capacity of louse larvae, which is sufficient to intercept a host over very small distances (millimetres to centimetres, as discussed in Reference [141]), may not be sufficient for most larvae to reach discrete trapping points or avoid an object the size of a commercial sea cage. Impregnation of cages with semiochemicals that either disrupt host-finding once larvae enter the cage (e.g., by blocking semiochemical receptors while lice are within the semiochemical plume) or attract lice to the mesh itself (including a method of trapping or killing larvae that are attracted to the mesh), are approaches that could be tested.

5.4 | Vaccines

Fish vaccination, which has been in use for over 40 years, has significantly contributed to the sustainability of the industry, and dramatically reduced the use of antibiotics.¹⁰ Most licensed vaccines currently used in aquaculture are produced using conventional methods and consist of inactivated or live-attenuated whole organisms, predominantly whole cell bacterins. In aquaculture, commercially available vaccines using modern technologies, targeting specific pathogen components, only include recombinant or subunit protein vaccines.¹⁰ Examples include an *E. coli*-based expression subunit vaccine against infectious pancreatic necrosis (IPN) in Norway (produced by Merck Animal Health) and a yeast-based subunit vaccine against infectious salmon anaemia (ISA) virus available in Chile (manufactured by Virbac-Centrovet).

Even though traditional vaccine technologies have enhanced live-stock productivity, the efficacy of inactivated vaccines can be suboptimal and live-attenuated ones may present safety concerns. To circumvent some of these flaws, veterinary medicine has been at the forefront of novel vaccine technology, pioneering the development and licensing of third-generation vaccines including DNA, RNA and recombinant viral-vector vaccines.⁶⁰⁸

The latest vaccine technology to be applied to aquaculture uses self-replicating RNA vaccines based on an alphavirus genome (i.e., salmonid alphavirus 3).⁶⁰⁹ Alphaviruses contain RNA replication machinery which is left intact in the vaccine, and genes encoding for structural proteins that are replaced with the antigen of interest. Such antigen-encoding RNA replicon platforms enable the production of a large amount of antigen from a small dose of vaccine. RNA vaccines have been found to give high protection against infectious salmon anaemia depending on the route of administration.^{610,611}

It is challenging to develop vaccines against parasites. In the case of ectoparasites, such as sea lice, there are even greater challenges

because of the antigenic complexity of metazoan organisms and physical separation between the pathogen and the host, allowing the pathogen to conceal a large proportion of its potential antigens. Despite these difficulties, and despite the few published research initiatives in this area (Table 6), researchers have recently demonstrated the protective effect of using a salmon lice-gut recombinant protein (P33) as a vaccine antigen against sea lice (*L. salmonis*) in a laboratory-based trial.⁶¹³ The identification of suitable antigens against sea lice followed a translational approach successfully implemented in the development of vaccines against ticks^{614,620} by proteomic identification of louse feeding-associated proteins. Under field conditions, a tick vaccine (Gavac; Heber Biotec), which is the only commercially available vaccine against an external parasite, uses a single tick gut antigen (Bm86) to induce protection against ticks (*Rhipicephalus microplus*) in cattle. The successful development of this tick vaccine, and the promising results observed in salmon immunised with the P33 vaccine have been achieved thanks to recent advances in ‘vaccinomics’, based on transcriptomic and proteomic data⁶²⁰ which allow a deeper understanding of the genetic factors and molecular pathways involved in the host–parasite interface. However, vaccines against lice may not be 100% effective, in which case they will not eliminate the lice problem but may reduce the number of lice per fish and/or the number of eggs produced per lice. This is the case with the tick vaccine for which 55%–100% efficacy in control was achieved 12–36 weeks after the first vaccination and 60% reduction in the number of acaricide treatments was achieved relative to that for non-vaccinated cattle.⁶²¹ If salmon lice vaccinations are not completely effective, animals may have to be revaccinated in the grow-out phase, which is a demanding and costly procedure as the fish grow larger, and other supplementary measures (such as biological control, medicines and selective breeding) will still be needed.

Transcriptomic profiling of differential responses to resistant and susceptible salmonid hosts has facilitated progress in understanding the importance of nutritional immunity in response to *Caligus rogercresseyi* infection and subsequent application of vaccination modulating iron-chelating activity.^{108,619} Injection of shrimp, *L. vannamei*, with recombinant ferritin has also demonstrated protective efficacy against WSSV by inhibiting viral replication.⁶²² Another recombinant approach which is more feasible to apply at large scale includes double-stranded RNA targeting VP28 (dsVP28) produced by the probiotic bacteria *Lactococcus lactis*. Administration of *L. lactis* expressing dsVP28 to the animals before the WSSV challenge, significantly increased survival and decreased viral load compared to non-treated animals.⁶²³ Differential infection-associated *L. salmonis* transcriptomic profiles between Atlantic and Pacific salmonid hosts has also revealed potential virulence genes,¹¹³ which may be exploited for vaccination.

Sequencing of homologous or conserved genes based on proteins and vaccine targets of ticks and other arthropods has proven an effective approach in antigen mining for sea lice.^{615–617} RNAi enables functional characterisation of vaccine candidates⁶²⁴ and has been applied to characterise a number of potential sea lice vaccine candidates including biomolecules involved in egg production (e.g., yolk-associated protein; LsYAP,⁶²⁵), digestion (e.g., KEDL receptor; LsKDELR and vesicular

coatmer protein complex; LsCOPB2,⁶²⁶), muscle activity (e.g., LsalMS,⁶²⁷) and immunomodulation (e.g., prostaglandin E-synthase 2; PGES2,⁶²⁸) leading to impaired parasite reproductive capacity, digestion and development. Gut digestion-associated serine proteases, for example, trypsin have been characterised based on sequence homology with other crustacea,⁶²⁹ secretory products such as metalloproteinases and collagenases have been identified by proteomics using LC-ESI-MS/MS,¹¹⁰ and exocrine glands associated with attachment and host–parasite interactions have been mapped and characterised microscopically.¹⁰⁷ Higher resolution sn-RNA-Seq and spatial- ‘omics’ technologies (Figure 1) will further elucidate the role glands and their secreted products play during louse infection, and these technologies are expected to result in the identification of new vaccine candidates.

6 | OPTIMISING IMPLEMENTATION AND DISSEMINATION TO ACHIEVE POPULATION-WIDE HOST RESISTANCE

The high cost of pathogen infection to the aquaculture industry is largely associated with the technology and labour involved with prevention, treatment and associated stresses, reduced growth and increased mortality thereafter (e.g., the high cost associated with delousing Atlantic salmon,^{27,36,630}). For strategies boosting host resistance to be effective preventative measures, we need to ensure that they eliminate or severely reduce the need for treatments such as delousing across the industry and are everlasting or sustainable.

While we can focus on the genetic basis of host resistance, it is important to have one eye on the broader population dynamics at play. There is little value, for example, in producing a host salmon or shrimp with twice the resistance if this does not translate into a dramatic reduction in actual infestation rates throughout the entire production cycle. Such nonlinear returns are entirely possible as implementation scales up from an individual to population level, especially for instance if the modification introduced can be repaired (as has been a problem when CRISPR gene drives are introduced into wild populations, for example,⁶³¹) or has little effect on the overall epidemiology of the disease. Alternatively, minor changes to resistance might have major consequences for infestation rates and may impose selection pressure on the parasite. Modelling of host–parasite dynamics, then, will provide an important tool for understanding how genetic manipulations might have maximum effect over a prolonged period. We focus here mainly on salmon louse as a case study. The ideas we discuss are, of course, relevant to the shrimp example also, but the shrimp example is more complex and critical factors (such as spatial connectivity) are not yet well resolved for WSSV.

6.1 | Understanding the epidemiological effects of genomic solutions

The full scope of genomic solutions to control infectious diseases in aquaculture is likely to be grossly under-realised because current

TABLE 6 Published sea lice vaccines and antigen discovery platforms

Vaccine antigens	Antigen localisation/ putative function	Antigen discovery technology	Administration method	Efficacy	References
Crude extract: Whole adult antigens	Gut antigen cocktail	Antigens characterised ConA enrichment and 1D SDS-PAGE/immunoblotting	Injection (intraperitoneal)	<i>Lepeophtheirus salmonis</i> 26% reduction in gravid female egg numbers.	[612]
<i>Escherichia coli</i> expressed recombinant protein: p33—potassium chloride amino acid transporter	Gut: Cellular hypotonic salinity response and transmembrane transportation	RP-LC-MS/MS proteomic identification of gut-associated proteins	Injection (intraperitoneal) with IP boost	<i>L. salmonis</i> 41.3% reduction in chalimus. 35.7% reduction in adult lice.	[613,614]
<i>E. coli</i> expressed recombinant protein: p30—putative toll-like receptor 6	Gut: Innate and adaptive immune response	RP-LC-MS/MS proteomic identification of gut-associated proteins	Injection (intraperitoneal) with IP boost	<i>L. salmonis</i> 31% reduction in chalimus. 16% reduction in egg string length.	[614]
<i>E. coli</i> expressed recombinant protein: pO ribosomal protein	Immunogenic midgut protein involved in assembly of 60S ribosomal subunit and protein synthesis	Sequencing data—low amino acid sequence similarity between louse and host (preventing tolerance or autoimmunity)	Injection (intraperitoneal) and immersion	<i>L. salmonis</i> 21% reduction of adult females, 42.5% reduction in gravid adult females. Delayed hatching of gravid female eggs, low copepodid counts in F1 generation.	[615,616]
<i>E. coli</i> expressed recombinant protein: my32-ls/akarín-like	Transcription factors required for NF- κ -dependent gene expression	Sequencing of conserved genes using degenerate primers and cloning	Injection (intraperitoneal)	<i>C. rogercresseyi</i> 57% reduction in adults.	[617,618]
<i>E. coli</i> expressed recombinant protein: IPATH [®] Iron binding domain of <i>S. salar</i> transferrin and <i>S. salar</i> ferritin subunit H	Iron-chelating – host blood iron regulatory proteins	Whole transcriptomic analysis/RNAseq using MiSeq Illumina ^(R) sequencing	Injection (intraperitoneal)	<i>C. rogercresseyi</i> 78% reduction of adults vs. 10% in controls. Disrupted embryogenesis, genital segment inflammation. Significant reduction in adult lice burden (mean 17 control vs. 407 in vaccinates)	[619]

selection does not consider host–parasite interactions and resulting epidemiological effects.⁶³² This is particularly pertinent in aquaculture programmes, in which disease resistance is defined as the ability to survive when exposed to infectious pathogens, as is the case for most virus infections, including WSSV in whiteleg shrimp.⁵⁸⁵ Survivors may have higher tolerance to cope with infections and thus transmit infections to others.⁷⁹ Few studies consider how endurance is affected by selection for disease resistance.²³⁴ Hence, it is generally not known whether and to what extent selecting directly on survival as the resistance phenotype reduces disease transmission and thus the incidence and severity of disease outbreaks in populations.⁶³³ Epidemiological models for micro-parasitic infections, such as virus infections, point to three underlying epidemiological host traits that affect pathogen transmission and subsequent mortality rates: susceptibility (the propensity of an uninfected individual to become infected when exposed to infectious material), infectivity (the ability of an individual, once infected, to transmit the infection) and mortality (the propensity of an

infected individual to die, i.e., the opposite of tolerance). Evidence from recent transmission experiments in aquaculture populations suggests that substantial host genetic variation and co-variation may exist for all three host traits, and that all three epidemiological traits can be reliably estimated.^{79,634} Furthermore, genetic-epidemiological models indicate that genomic selection for reduced host susceptibility and infectivity may more effectively reduce disease prevalence than current selection for disease resistance.⁶³⁵

Genetic selection for host resistance to external-parasites like sea lice is expected to directly reduce parasite load in the populations, as observed in practice for sea lice infections in salmon.⁴⁶ This is because parasite count is used as the resistance phenotype. For example, resistant fish with lower sea lice counts are expected to also propagate fewer lice and hence reduce the overall lice prevalence in the population.⁶⁰ Nevertheless, the full potential of genomics is also likely to be under-realised for these types of infections. There are many host resistance mechanisms that interfere with the parasite life cycle that

are likely under host genetic control and could therefore be targeted for genetic improvement. These include, for example, those that lead to (a) an increasing proportion of non-gravid and thus non-reproductive females due to a reduction in the average lice density per fish⁶³⁶; (b) reduced fertility of the gravid female lice; (c) lower hatchability of the eggs; (d) lower survival from hatching to the infective copepodid stage; (e) lower infective success rate of the copepodids; (f) lower survival success of copepods through behavioural divergence or lower quality energetic resources. Factors (b)–(f) may be caused by lice maturing on fish with increased resistance to lice thus leading to an inferior rearing environment due to, for example, different chemical factors in the mucus and skin of the salmon host. This is expected to result in a higher genetic gain in resistance to the lice than predicted from classical quantitative genetic theory. Research is needed to assess the host genetic effects underlying such mechanisms. In the first instance for the lice example, it would be useful to assess the differences in the number of male lice and the number of gravid and non-gravid female lice, number of eggs per female lice, and infectivity of lice growing on fish with very high and low estimated breeding values for lice density.

A better understanding of the influence of host resistance on parasite epidemiology will allow the development of more efficient strategies to reduce lice infestation, including evaluation of the economic importance of performing genetic improvement for increased resistance of the host to the parasite and formulation of functional feeds. In particular, such data would inform epidemiological models of parasite prevalence in host populations that differ genetically in terms of their impact on the parasite life-cycle.⁶⁰ Such models are needed for optimising selective breeding strategies and furthering our understanding of the effect of selection for increased parasite resistance on possible correlated epidemiological effects through reduced parasite reproduction.

6.2 | Mitigating possible counter-evolution by the parasite or pathogen

It is well known that parasites and pathogens can rapidly adapt to pest controls used on farms.^{637–640} Without proper precautions in place, pest evolution can lead to a dramatic decline in the efficacy of a novel control technology. The Atlantic population of *L. salmonis* has already evolved pesticide resistance to most of the chemical treatments used on salmon farms,^{48,641} and there are concerns that they could similarly adapt to the various non-chemical alternatives that are in use.⁴⁷ Due to the strong louse gene flow between farms, advantageous traits can spread through the parasite population in a very short space of time.^{641–643} When developing new, parasite-resistant strains of host, it is therefore imperative that the risk of counter-adaptations evolving in the parasite is fully assessed.

Just as individual Atlantic salmon vary in their ability to resist lice,^{62,188} so too may there be genetic variation within *L. salmonis* in their ability to tolerate, evade or modulate the hosts' immune defences. Lice with improved infestation success and survival on

resistant salmon strains would therefore be selected for. The possibility for interaction effects between salmon and louse genotypes on infestation success is yet to be studied.

In terrestrial agriculture, pathogens have evolved to overcome resistant genes in plants on many occasions.^{644–647} Increased use of transgenic crops that have been modified to produce insecticidal proteins has driven a surge of counter-adaptations in insect pests.⁶⁴⁰ However, selective breeding for resistance in animals has proven to be much more durable against counter-adaptations, providing long-standing protection against parasites.^{648,649} Nevertheless, host–parasite co-evolutionary arms races occur in many animal systems,^{650–653} and so the potential for lice to adapt to changes in host resistance must be considered. Theoretical studies on parasite adaptation in hosts with different genetic resistance to the parasite suggest that it is unlikely that the parasite would change significantly as a result of selective breeding for increased parasite resistance.⁶⁵⁴ This is explained by the fact that adaptation of the pathogen requires allele(s) that are favourable in hosts with improved resistance, that long-term selective breeding for increased disease resistance is a dynamic and broad spectra strategy involving very many genes and that the resilience in the host is rarely complete, varies between fish, and the genetic progress is relatively small per generation of breeding. This is why strategies involving selective breeding for host resistance to lice are probably less risky than, for example, drugs or vaccines that affect one or a few specific mechanisms of interaction between the host and the pathogen.⁶⁴⁸

Lepeophtheirus salmonis populations in the Atlantic and Pacific Oceans belong to two allopatric subspecies that likely diverged ~5 million years ago.^{655,656} Given that parasite evolution is profoundly shaped by the host environment, one would expect the Atlantic and Pacific louse subspecies to have locally adapted to their sympatric *Salmo* and *Oncorhynchus* hosts, respectively, to some degree over these ~5 million years. This topic has received limited research but will be relevant if resistance mechanisms in Pacific salmon species are used as the template for gene-edited Atlantic salmon. That Pacific species such as coho salmon have retained their relatively high resistance, despite millions of years of co-evolution with lice, suggests that lice will be unlikely to overcome coho genes used as the basis for gene edits to produce resistant Atlantic salmon.⁶⁴⁸ Nevertheless, the full efficacy of this strategy could erode over time through local adaptations in the louse population. In the wild, coho salmon can still support relatively high louse infestations.^{657–659}

Pacific salmon lice show an enhanced transcriptomic feeding response on Atlantic salmon compared with on Pacific hosts.¹¹³ Louse responses on sympatric coho and sockeye salmon were similar, despite these host species differing in resistance.¹¹³ Fast et al.¹⁰⁰ measured the production of enzymes secreted by Pacific and Atlantic lice, thought to assist in digestion and/or host immunomodulation, in response to the mucus of different host species. Both louse subspecies had higher enzyme production when exposed to the mucus of Atlantic salmon, than when exposed to more resistant coho salmon (*O. kisutch*). Interestingly, mucus tended to stimulate a stronger feeding response in sympatric, rather than allopatric, louse subspecies.¹⁰⁰

That is, Atlantic salmon mucus and coho mucus stimulated relatively higher enzyme secretion in Atlantic and Pacific lice, respectively. These differences in the feeding activity of lice on sympatric and allopatric host species^{100,113} highlight the heightened susceptibility of Atlantic salmon to lice and are also suggestive of some degree of local adaptation.

Research into infestations on non-salmonid hosts, such as the three-spined stickleback (*Gasterosteus aculeatus*), may also provide insights into the extent to which lice can adapt to alternative host strains. *Lepeophtheirus salmonis* will readily infest sticklebacks in the laboratory and the wild, but do not appear to be able to successfully sexually mature on them.^{660,661}

Given the effort required to research and develop new mechanisms of host resistance, it is desirable for a strategy to maintain a high level of efficacy for as long as possible. A key plank of any plan for implementing genetic technologies for host resistance should be to minimise the risk of counter-adaptation by the parasite. An evolutionarily stable strategy consistent with animal welfare and production demands is therefore needed. One approach for achieving this is to maintain genetic diversity in host resistance, both within and between host strains. The more genes for resistance that are incorporated into a host strain, the more complex the mechanism of resistance, and the harder it is for parasites to evolve counter-adaptations.^{662–664} Of course, the trade-off of this is the extra effort required to develop additional gene edits and focus selective breeding on disease resistance while maintaining high levels of genetic diversity in the captive-bred populations.

Another approach for slowing the spread of parasite counter-adaptation might be to establish refugia—in the salmon example, maintaining populations of susceptible salmon strains.^{640,665,666} Refugia are particularly effective at preventing parasite adaptation if counter-adaptations incur fitness costs on susceptible host strains.^{666,667} Wild populations can act as refugia, provided they are large enough.^{665,668} In the Atlantic, however, farmed salmon are significantly more abundant than wild hosts.³⁴ It might be more effective, then, if some farms were to act as refugia instead, by being stocked with susceptible host strains, but it seems unlikely that such deliberate degradation of fish welfare on a whole-farm basis would be acceptable to the authorities and public.

Choosing which farms must forego resistant hosts to avoid counter-adaptations would pose a difficult decision, as would decide how many different gene edits are needed to provide an evolutionarily durable strategy. This is where evolutionarily dynamic metapopulation models can be powerful tools (e.g.,⁶⁶⁹). Such models have previously been used to predict the rapid spread of pesticide resistance, and to identify evolutionary hotspots in areas of intensive farming.⁶⁶⁹ To simulate counter-adaptation to resistant hosts, these models could include the stocking of different host strains, selection imposed by these strains on louse genotypes, and recombination and mutation of louse genes. Model simulations can then be run for different scenarios to determine the optimal management regime: one that prevents lice from adapting to resistant hosts, while still significantly reducing lice infestations in the short term.

These models are valuable for identifying the most evolutionarily durable avenues for achieving host resistance. Once resistant strains have been developed, models can again be used, this time to inform how resistant salmon should be introduced to farms through space and time. Simulations can also be used to consider how resistant salmon should be deployed in concert with other preventative strategies,⁴⁶ to further mitigate the risk of louse evolution.⁶⁷⁰ Of course, the accuracy of such models is directly proportional to our understanding of counter-adaptations in lice. The more that is known about the mechanisms, genetic architecture and fitness costs behind any counter-adaptations, the greater the predictive power of the model.

6.3 | Integration of genomic technologies and dissemination to the sector with the implementation of gene editing and genomic selection

Genome editing in combination with other biotechnological advancements such as germ cell technologies could be used to help accelerate genetic improvement.^{599,671} Gene editing can rapidly introduce favourable changes to the genome, either by fixing alleles at existing trait loci, creating de novo alleles or introducing alleles from other strains or species. However, seamless integration of genome editing technologies into a well-managed breeding programme is required to ensure continuous genetic improvement and careful management of genetic diversity. It is also important to consider that gene editing is likely to alter the genetic architecture of other traits of interest, perhaps leading to substantial epistasis, and this would have to be accounted for in future breeding strategies.

The expected genetic improvement of traits of interest through selective breeding depends on several factors including heritability, selection intensity, selection accuracy and generation interval. The implementation of genome editing in aquaculture breeding will have the same goals as traditional selective breeding practices. In the cases here, the ultimate goals for genetic improvement should be to eliminate or reduce the number of delousing events required by the salmon industry and to boost the survival of shrimp in the face of WSSV disease. The number of lice that have infected the fish in challenge and/or field tests are the index traits that are being used for the salmon lice case whereas survival after a challenge test is the index trait currently used to select for WSSV resistance. However, host resistance is a complex phenotype consisting of multiple layers (attraction, prevention of attachment or infection, immune response, infectiousness, etc.). More detailed knowledge about host influences on disease reproductive success, host immunology and host semiochemicals influencing lice attraction and attachment could provide more specific phenotypes for host selection and enable more accurate forms of genomic selection for host resistance. Such knowledge will also provide targets for gene editing.

The effect of gene editing on disease resistance could be on a similar scale to that of a major gene affecting a trait, or in the case of a de novo edit, could be on a much larger scale (effecting disease resistance to an extent not seen within the breeding population). If

the gene edits were directed at genetic variants causing QTL effects (in a way that creates similar variants to those that naturally exist in the population), the editing could have a potentially large impact on traits where one or a few major genes affect the trait considerably. In this case, the gene editing might bring us a large step forward by fixing a preferred allele for a gene underlying a QTL. For traits with a more polygenic structure, traditional selection and genomic selection would be preferred to this type of gene editing in most instances. In any case, the application of selection methods will improve the trait further, by selectively improving the genotypes at other loci related to the same trait.

However, as noted above, editing may alter the genetic architecture of the trait and lead to substantial epistasis. In plants, gene editing has been suggested as a final step after selection for complex traits, to improve monogenic quality traits.⁶⁷² In aquaculture, gene editing needs to be done before selection for other traits has occurred. The edited fish might not therefore be chosen as a parent if they have poor performance for other traits of interest. Alternatively, surrogate broodstock technologies⁵⁹⁹ could help to reduce the long generation interval in Atlantic salmon and be used as a dissemination tool because the editing could be implemented after selection and before dissemination to producers without altering the breeding programme germplasm. The costs associated with these strategies (increased direct costs and/or reduced gain for other traits) may be acceptable if the gene editing is successful, important and does not need to be repeated every generation. However, if editing needs to be performed repeatedly, to cover many genes or traits, a more integrated approach is needed where for instance a few edited and selected individuals are used to introgress the edited variant into the population.

Editing efficiency also determines the feasibility of gene editing in aquaculture breeding. Studies on the optimal number of edited animals required in aquaculture breeding schemes are lacking, but simulation results from livestock schemes demonstrated that genome editing technologies in combination with genomic selection have the potential to increase genetic gain many-fold for traits of interest compared with genomic selection alone.^{673–675} As of today, no cost-benefit analysis has been done for gene-edited assisted selection (GEAS) programmes in aquaculture. Such an analysis would consider the number of gene-edited individuals, single or multiple gene edit effects, other traits under selection, expected genetic gain and rate of inbreeding. Also, the rate of dissemination and the economic implications of these schemes should be assessed and compared with conventional breeding designs. Specific GEAS schemes with special attention to multiplier parents, which produce seed for the production tier, could then be designed. The multiplier parents carrying resistance edited genes to be selected, and the effect of this selection strategy on the resistance of the production population, should be investigated. A recent modelling study revealed that gene editing, when effectively combined with other disease control strategies, such as vaccination, may be able to eliminate persistent livestock diseases that are currently difficult to control.⁶⁷⁶ However, according to this study, such desirable outcomes can only be achieved if the distribution of

genetically resistant individuals into commercial populations is coordinated by breeding companies or national schemes.

7 | ETHICAL CONSIDERATIONS

Improving disease resistance improves the economics of aquaculture, and it also contributes to more sustainable aquaculture production by improving animal welfare and reducing the impact of aquaculture on wild stocks.⁶⁷⁷ Traditionally, selection for genetic improvement is based on survival information from challenge-tested siblings of the breeding candidates, which involves exposing large numbers of individuals to pathogens, and therefore results in some suffering. Survival phenotypes are recorded from numerous relatives to enable accurate estimation of breeding values. This sacrifice can be argued to be ethically acceptable as it is done for avoiding suffering of a much greater number of individuals in large-scale disease outbreaks. Even so, the intrinsic value and welfare of fish, independent of their utility to humans, should be part of our ethical considerations.⁶⁷⁷ Adoption of advanced breeding strategies (e.g., genomic selection) and/or adoption of new biotechnological methods (e.g., gene editing) will reduce the use of experimental animals. However, other ethical dilemmas arise when technologies such as gene editing are proposed to be adopted and implemented in practical breeding programmes.

Knowledge particular to each case is needed so that we can understand and evaluate potential off-target and/or pleiotropic effects of gene editing on welfare. Another important consideration will be how the edits should be most effectively and responsibly created and spread throughout the breeding and multiplier populations. To ensure sustainable implementation of gene editing in aquaculture it is important to ensure that there is no possibility for edited fish to detrimentally affect wild population gene pools. The effect of escapees from a gene-edited population into the wild is not only dependent on the number of escaped fish, but also their phenotypic characteristics related to fitness and the existing diversity in the receiving ecosystem.⁶⁷⁸ Genetic introgression from aquaculture to the wild populations could be avoided by making the gene-edited fish sterile (either through editing of a locus affecting fertility or other means). In summary, the application of gene editing to aquaculture needs to be safe for the fish, the consumer and the environment⁶⁷⁹ and every proposed edit needs to be researched and evaluated with risk analyses on a case-by-case basis.

Research institutes producing gene-edited fish and shellfish need to ensure future sustainable and beneficial impacts from the use of gene editing by developing strict guidelines for a *Responsible Research and Innovation* (RRI) framework. RRI frameworks commit to (1) describe, analyse and openly discuss the consequences of the process and outcomes of the research activities (anticipation), (2) holistically reflect on the moral, political and social assumptions and the main motivation and drivers of the research in question (reflexivity), (3) invite all relevant stakeholders to contribute views and opinions about the research trajectory (inclusion) and (4) be willing to change the trajectory of research if feedback from the stakeholders reflects that

research is not likely to meet the requirement and expectations of the society (responsiveness).⁶⁸⁰ Rigorous quality systems need to be set in place to ensure that such guidelines are followed and that the fish and shellfish produced are securely contained.

Due to the complexity of the biotechnology, fish biology and of each specific situation, a case-by-case comprehensive and systematic approach is needed to address the ethical acceptability of each technological advance.^{681,682} Ethical matrix (EM) approaches rely on generally accepted ethical principles and are used to get an overview of the most important ethical interests or values connected to the particular issue.⁶⁸¹ The formulation of the overview involves and considers all groups who have an interest in or are affected by the technology including industry, NGO and government, and considers effects on the individual animals that are directly targeted, as well as the overall effect on nature. Emphasising the principles of well-being, justice and fairness, the EM was designed to create discussion over ethical prioritisation for those with little or no training in ethical theory. EM can be used as a tool both to guide the research process within a project group and to create a dialogue between scientists and stakeholders. EM approaches can be used repeatedly and involve different stakeholder groups to promote the inclusiveness and responsiveness of the research project.

Any dissemination plan produced should follow and expand on institutional RRI guidelines that have developed for the use of gene editing (e.g.,⁶⁸⁰). Following the RRI dimension on inclusion, data should be shared and workshops held involving diverse scientific and industry representatives so that insight is gained, tapping into broad interdisciplinary knowledge and practical farming experiences. Furthermore, industry partners should be consulted at every step and be represented on the steering committee for such projects. Methodological ethical considerations include, for example, performing a priori power calculations to guarantee adequate but not unnecessarily large experimental setups, execution of challenge tests in bio-secure environments to prevent pathogenic, parasitic, or edited material from escaping en masse into the surrounding environment, close monitoring of experimental animals and euthanasia of suffering individuals. In addition to using breeding strategies and technological methods that minimise challenge tests, more focus should be put towards assessment of alternative less invasive phenotypes (as an alternative to exposing fish to pathogens in challenge test).

With the work that is underway around the world applying genetic technologies to combat infectious diseases in aquaculture, aquaculture sectors will soon face important decisions. Likely there will be a much greater understanding of natural processes affecting host resistance to parasites and pathogens and strategies will be developed that could transform these industries as we know them. The projects now underway will advise on the most responsible and effective approach, or combination of approaches, for reducing or eliminating these parasite and disease problems. There will be a need for further testing of gene editing to ensure there are no risks. The aquaculture sectors involved need to have an open dialogue around the use of these technologies. 'Business as usual', with no change, is not a viable option, considering the effects of these diseases on

profitability, animal welfare and public perceptions. The goal of such research will be to develop safe and effective strategies. But it is the aquaculture industry sectors involved that will need to work with relevant public and government agencies to decide if these strategies should be implemented, and to clear the path, so that they can solve this problem and transform their businesses.

8 | CONCLUSIONS

Substantial research programmes are underway that aim to produce new knowledge that could be applied for boosting host resistance to eliminate or severely reduce infections by, for instance, sea lice in salmon and WSSV in shrimp. These projects are utilising a suite of technologies that have been enabled by ultra-high-throughput sequencing, such as single nuclei and spatial transcriptomics and SNP GWAS. Newly developed methodologies like in-vivo or in-vitro gene editing and functional testing hold great promise for helping to find and test genetic mechanisms affecting host resistance. These projects are also exploring the possibility of using genomic selection and gene editing with CRISPR-Cas9 to create host populations that will resist these diseases. The implementation of these technologies needs to be carefully considered. Practical methods that will allow easy adoption, implementation and dissemination by aquaculture sectors are needed. Population genetic variability needs to be maintained, inbreeding limited and possibilities for the genetic improvement of other important traits must be ensured. Ethical concerns, particularly about the use of gene editing methodology, need to be openly discussed and debated in public arenas, and thorough testing and safeguards (e.g., sterilisation) are needed to ensure that there are no negative consequences for the wild populations of these species or for the broader ecosystem.

The application of new genomic technologies and methodologies is expected to generate knowledge about genes that trigger a more effective immune response in some species or lines; the effect that could be realised by editing these genes in more susceptible species or lines; potential lice attractants, repellents and assays; and the extent of additive genetic variation affecting the production and release of important immune factors and semiochemicals. Such knowledge could lead to the development of feed additives, gene edits, new vaccines and the enhancement of genomic breeding value estimation to promote host resistance. The epidemiological implications of these applications on the infectivity and virulence of aquatic diseases needs to be explored, and routines need to be devised to enhance the suppression of disease in the general aquaculture environment.

Such projects are ambitious in that it is hypothesised that specific semiochemical or immune pathways play major roles in differentiating disease-resistant from disease-susceptible hosts and that these differences are measurable, have a strong genetic basis, have implications for the epidemiology of infection and that genomic selection and/or gene editing approaches can be effectively and sustainably applied to reduce or eliminate the effect of disease on the host without counter evolutionary responses by the infectious agent taking effect. The

long-term suppression of disease will only be realised through a collaborative and coordinated multi-disciplinary effort involving scientists working closely with aquaculture industry and government. Such efforts are likely to significantly advance our understanding of host-parasite and host-disease interactions and mechanisms affecting resistance to disease and should result in significant economic impacts for aquaculture sectors, benefit the welfare of production animals and create ecosystem benefits for natural populations of these species.

AUTHOR CONTRIBUTIONS

Diego Robledo: Conceptualization; funding acquisition; investigation; methodology; visualization; writing – original draft; writing – review and editing. **Lene Sveen:** Conceptualization; funding acquisition; investigation; methodology; visualization; writing – original draft; writing – review and editing. **Rose Ruiz Daniels:** Investigation; methodology; writing – original draft; writing – review and editing. **Aleksei Krasnov:** Conceptualization; funding acquisition; investigation; methodology; writing – original draft; writing – review and editing. **Andrew Lindsay Coates:** Investigation; methodology; writing – original draft; writing – review and editing. **Ye Hwa Jin:** Investigation; writing – original draft; writing – review and editing. **Luke Barrett:** Writing – original draft; writing – review and editing. **Marie Lillehammer:** Formal analysis; writing – original draft; writing – review and editing. **Anne Helena Kettunen:** Methodology; writing – original draft; writing – review and editing. **Ben L Phillips:** Conceptualization; funding acquisition; investigation; writing – original draft; writing – review and editing. **Tim Dempster:** Conceptualization; investigation; methodology; supervision; writing – original draft; writing – review and editing. **Andrea Doeschl-Wilson:** Formal analysis; writing – original draft; writing – review and editing. **Francisca Samsing:** Writing – original draft; writing – review and editing. **Gareth Difford:** Investigation; writing – original draft; writing – review and editing. **Sarah Salisbury:** Investigation; writing – original draft; writing – review and editing. **Bjarne Gjerde:** Conceptualization; investigation; methodology; writing – original draft; writing – review and editing. **John-Erik Haugen:** Investigation; writing – original draft; writing – review and editing. **Erik Burgerhout:** Methodology; writing – original draft; writing – review and editing. **Binyam Dagnachew:** Investigation; writing – original draft; writing – review and editing. **Dominic Kurian:** Investigation; writing – original draft; writing – review and editing. **Mark D. Fast:** Conceptualization; funding acquisition; methodology; visualization; writing – original draft; writing – review and editing. **Morten Rye:** Conceptualization; funding acquisition; writing – original draft; writing – review and editing. **Marcela Salazar:** Investigation; writing – original draft; writing – review and editing. **James Bron:** Conceptualization; funding acquisition; methodology; writing – original draft; writing – review and editing. **Sean Monaghan:** Investigation; writing – original draft; writing – review and editing. **Celeste Jacq:** Funding acquisition; investigation; writing – original draft; writing – review and editing. **Mike Birkett:** Funding acquisition; investigation; writing – original draft; writing – review and editing. **Howard Browman:** Funding acquisition; investigation; writing – original draft; writing – review and editing. **Anne Berit Skiftesvik:** Investigation; writing – original draft;

writing – review and editing. **David Fields:** Investigation; writing – original draft; writing – review and editing. **Erik Selander:** Investigation; writing – original draft; writing – review and editing. **Samantha Bui:** Investigation; writing – original draft; writing – review and editing. **Anna Sonesson:** Writing – original draft; writing – review and editing. **Stanko Skugor:** Methodology; writing – original draft; writing – review and editing. **Tone-Kari Knutsdatter Østbye:** Conceptualization; funding acquisition; investigation; methodology; writing – original draft; writing – review and editing. **Ross Houston:** Conceptualization; funding acquisition; investigation; supervision; writing – original draft; writing – review and editing.

ACKNOWLEDGEMENTS

This review was funded from our projects with the short titles ‘CrispResist’ funded by the Norwegian Seafood Research Fund (FHF, project 901631), ‘NoLice’ and GenomResist’ funded by the Norwegian Research Council (projects 320619 and 244131 respectively) and ‘GenoLice’ funded by the Biotechnology and Biological Sciences Research Council of the UK. We also thank our industry partners Benchmark Genetics, Mowi, Salmar, the Sustainable Aquaculture Innovation Centre, Centre for Aquaculture Technologies Canada and Cargill for their contributions of knowledge, time, animals and resources to these projects.

DATA AVAILABILITY STATEMENT

Data sharing not applicable - no new data generated.

ORCID

Nicholas A. Robinson  <https://orcid.org/0000-0003-1724-2551>
 Diego Robledo  <https://orcid.org/0000-0002-9616-5912>
 Lene Sveen  <https://orcid.org/0000-0003-3394-3462>
 Rose Ruiz Daniels  <https://orcid.org/0000-0002-6702-5304>
 Andrew Coates  <https://orcid.org/0000-0002-6455-7372>
 Luke T. Barrett  <https://orcid.org/0000-0002-2820-0421>
 Marie Lillehammer  <https://orcid.org/0000-0001-5092-1105>
 Anne H. Kettunen  <https://orcid.org/0000-0002-1217-7079>
 Tim Dempster  <https://orcid.org/0000-0001-8041-426X>
 Andrea Doeschl-Wilson  <https://orcid.org/0000-0002-2658-6973>
 Francisca Samsing  <https://orcid.org/0000-0002-6343-2295>
 Erik Burgerhout  <https://orcid.org/0000-0002-5737-3575>
 Binyam S. Dagnachew  <https://orcid.org/0000-0002-5166-5497>
 James E. Bron  <https://orcid.org/0000-0003-3544-0519>
 Sean J. Monaghan  <https://orcid.org/0000-0002-7692-7756>
 Samantha Bui  <https://orcid.org/0000-0002-7885-2989>
 Ross D. Houston  <https://orcid.org/0000-0003-1805-0762>

REFERENCES

1. Anonymous. Harness the world's aquatic ‘blue’ food systems to help end hunger. *Nature*. 2021;597(7876):303.
2. Jahangiri L, MacKinnon B, St-Hilaire S. Infectious diseases reported in warm-water marine fish cage culture in East and Southeast Asia—a systematic review. *Aquac Res*. 2022;53:2081-2108.
3. Oidtman B, Dixon P, Way K, Joiner C, Bayley AE. Risk of waterborne virus spread—review of survival of relevant fish and

- crustacean viruses in the aquatic environment and implications for control measures. *Rev Aquacult.* 2018;10(3):641-669.
4. Fan YJ, Chan KH, Hung IFN. Safety and efficacy of COVID-19 vaccines: a systematic review and meta-analysis of different vaccines at phase 3. *Vaccine.* 2021;9(9):989.
 5. Adu-Bobie J, Capecchi B, Serruto D, Rappuoli R, Pizza M. Two years into reverse vaccinology. *Vaccine.* 2003;21(7-8):605-610.
 6. Bogwald J, Dalmo RA. Review on immersion vaccines for fish: an update 2019. *Microorganisms.* 2019;7(12):627.
 7. Dalmo RA. DNA vaccines for fish: review and perspectives on correlates of protection. *J Fish Dis.* 2018;41(1):1-9.
 8. Evelyn TPT. A historical review of fish vaccinology. In: Gudding R, Lillehaug A, Middlyng P, Brown F, eds. *Fish Vaccinology.* Vol 90; Basel, Switzerland: Karger Postfach; 1997:3-12.
 9. Ji QY, Wang SY, Ma JF, Liu Q. A review: Progress in the development of fish vibrio spp. vaccines. *Immunol Lett.* 2020;226:46-54.
 10. Ma J, Bruce TJ, Jones EM, Cain KD. A review of fish vaccine development strategies: conventional methods and modern biotechnological approaches. *Microorganisms.* 2019;7(11):569.
 11. Mohd-Aris A, Muhamad-Sofie MHN, Zamri-Saad M, Daud HM, Ina-Salwany MY. Live vaccines against bacterial fish diseases: a review. *Vet World.* 2019;12(11):1806-1815.
 12. Tonheim TC, Bogwald J, Dalmo RA. What happens to the DNA vaccine in fish? A review of current knowledge. *Fish Shellfish Immunol.* 2008;25(1-2):1-18.
 13. Mariner JC, House JA, Mebus CA, et al. Rinderpest eradication: appropriate technology and social innovations. *Science.* 2012;337(6100):1309-1312.
 14. Schleiff M, Olateju A, Decker E, et al. A multi-pronged scoping review approach to understanding the evolving implementation of the smallpox and polio eradication programs: what can other Global Health initiatives learn? *BMC Public Health.* 2020;20:1698.
 15. Willadsen P. Vaccination against ectoparasites. *Parasitology.* 2006;133:S9-S25.
 16. Arala-Chaves M, Sequeira T. Is there any kind of adaptive immunity in invertebrates? *Aquaculture.* 2000;191:247-258.
 17. FAO. FAO Global Fishery and Aquaculture Production (Version 2021.1.2). In: Nations FaAOotU, 2021.
 18. Costello MJ. Ecology of sea lice parasitic on farmed and wild fish. *Trends Parasitol.* 2006;22(10):475-483.
 19. Hamre LA, Eichner C, Caipang CMA, et al. The Salmon louse *Lepeophtheirus salmonis* (Copepoda: Caligidae) life cycle has only two Chalimus stages. *PLoS One.* 2013;8(9):e73539.
 20. Brooker AJ, Skern-Mauritzen R, Bron JE. Production, mortality, and infectivity of planktonic larval sea lice, *Lepeophtheirus salmonis* (Krøyer, 1837): current knowledge and implications for epidemiological modelling. *ICES J Marine Sci.* 2018;75(4):1214-1234.
 21. Ho JS, Kim IH, Cruz-Lacierda ER, Nagasawa K. Sea lice (Copepoda, Caligidae) parasitic on marine cultured and wild fishes of The Philippines. *J Fisheries Soc Taiwan.* 2004;31(4):235-249.
 22. Nagasawa K. Sea lice, *Lepeophtheirus salmonis* and *Caligus orientalis* (Copepoda: Caligidae), of wild and farmed fish in sea and brackish waters of Japan and adjacent regions: a review. *Zool Stud.* 2004;43(2):173-178.
 23. Tanaka S, Yamamoto S, Ogawa K. The occurrence of *Caligus sclerotinosus* (Caligidae) infection in cultured Red Sea bream *Pagrus major* and involvement of Phototaxis in fish-to-fish transfer of the adults. *Fish Pathol.* 2013;48(3):75-80.
 24. Hvas M, Bui S. Energetic costs of ectoparasite infection in Atlantic salmon. *J Exp Biol.* 2022;225(1):jeb24330.
 25. Øverli Ø, Nordgreen J, Mejdell CM, et al. Ectoparasitic sea lice (*Lepeophtheirus salmonis*) affect behavior and brain serotonergic activity in Atlantic salmon (*Salmo salar* L.): perspectives on animal welfare. *Physiol Behav.* 2014;132:44-50.
 26. Fjelldal PG, Hansen TJ, Karlsen Ø. Effects of laboratory salmon louse infection on osmoregulation, growth and survival in Atlantic salmon. *Conserv Physiol.* 2020;8(1):coaa023.
 27. Overton K, Dempster T, Oppedal F, Kristiansen TS, Gismervik K, Stien LH. Salmon lice treatments and salmon mortality in Norwegian aquaculture: a review. *Rev Aquacu.* 2019;11(4):1398-1417.
 28. Overton K, Oppedal F, Stien LH, Moltumyr L, Wright DW, Dempster T. Thermal delousing with cold water: effects on salmon lice removal and salmon welfare. *Aquaculture.* 2019;505:41-46.
 29. Overton K, Samsing F, Oppedal F, Dalvin S, Stien LH, Dempster T. The use and effects of hydrogen peroxide on salmon lice and post-smolt Atlantic salmon. *Aquaculture.* 2018;486:246-252.
 30. Costello MJ. How sea lice from salmon farms may cause wild salmonid declines in Europe and North America and be a threat to fishes elsewhere. *Proc Roy Soc Biol Sci.* 2009;276:3385-3394.
 31. Taranger GL, Karlsen Ø, Bannister RJ, et al. Risk assessment of the environmental impact of Norwegian Atlantic salmon farming. *ICES J Mar Sci.* 2014;72(3):997-1021.
 32. Kristoffersen AB, Qviller L, Helgesen KO, Vollset KW, Viljugrein H, Jansen PA. Quantitative risk assessment of salmon louse-induced mortality of seaward-migrating post-smolt Atlantic salmon. *Epidemics.* 2018;23:19-33.
 33. Krkošek M, Lewis MA, Volpe JP. Transmission dynamics of parasitic sea lice from farm to wild salmon. *Proc Roy Soc B: Biol Sci.* 2005;272(1564):689-696.
 34. Dempster T, Overton K, Bui S, et al. Farmed salmonids drive the abundance, ecology and evolution of parasitic salmon lice in Norway. *Aquac Environ Interact.* 2021;13:237-248.
 35. Vollset KW, Krontveit RI, Jansen PA, et al. Impacts of parasites on marine survival of Atlantic salmon: a meta-analysis. *Fish Fish.* 2016;17(3):714-730.
 36. Iversen A, Hermansen Ø, Nystøyl R, Rolland KH, Garshol LD. Konkurransenevne for norsk oppdrettslaks: Kostnader og kostnadsdrivere i Norge og konkurrentland. *Nofima Rapport.* 2019;28:1-77.
 37. Abolofia J, Asche F, Wilen JE. The cost of lice: quantifying the impacts of Parasitic Sea lice on farmed Salmon. *Mar Resour Econ.* 2017;32(3):329-349.
 38. Godwin SC, Fast MD, Kuparinen A, Medcalf KE, Hutchings JA. Increasing temperatures accentuate negative fitness consequences of a marine parasite. *Sci Rep.* 2020;10(1):18467.
 39. Sandvik AD, Dalvin S, Skern-Mauritzen R, Skogen MD. The effect of a warmer climate on the salmon lice infection pressure from Norwegian aquaculture. *ICES J Mar Sci.* 2021;78(5):1849-1859.
 40. Medcalf KE, Hutchings JA, Fast MD, Kuparinen A, Godwin SC. Warming temperatures and ectoparasitic sea lice impair internal organs in juvenile Atlantic salmon. *Mar Ecol Prog Ser.* 2021;660:161-169.
 41. Hemmingsen W, MacKenzie K, Sagerup K, Remen M, Bloch-Hansen K, Imsland AKD. *Caligus elongatus* and other sea lice of the genus *Caligus* as parasites of farmed salmonids: a review. *Aquaculture.* 2020;522:735160.
 42. Boxshall GA, Bravo S. On the identity of the common *Caligus* (Copepoda: Siphonostomatoida: Caligidae) from salmonid netpen systems in southern Chile. *Contrib Zool.* 2000;69(1-2):137-146.
 43. Bravo S, Erranz F, Lagos C. A comparison of sea lice, *Caligus rogercresseyi*, fecundity in four areas in southern Chile. *J Fish Dis.* 2009;32(1):107-113.
 44. Bravo S, Silva MT, Treasurer J. Factors affecting the abundance of *Caligus rogercresseyi* (Boxshall and Bravo) on farmed salmonids in Chile in the period 2006-2007. *Aquaculture.* 2014;434:456-461.
 45. Liu YJ, Bjelland HV. Estimating costs of sea lice control strategy in Norway. *Prev Vet Med.* 2014;117(3-4):469-477.
 46. Barrett LT, Oppedal F, Robinson NA, Dempster T. Prevention not cure: a review of methods to avoid sea lice infestations in salmon aquaculture. *Rev Aquac.* 2020;12(4):2527-2543.

47. Coates A, Phillips BL, Bui S, Oppedal F, Robinson NA, Dempster T. Evolution of salmon lice in response to management strategies: a review. *Rev Aquac.* 2021;13(3):1397-1422.
48. Aaen SM, Helgesen KO, Bakke MJ, Kaur K, Horsberg TE. Drug resistance in sea lice: a threat to salmonid aquaculture. *Trends Parasitol.* 2015;31(2):72-81.
49. Gonzalez RR, Himelreichs JF, Cruzat FA, Asencio GC, Oyarzun P, Hernandez-Miranda E. Frequent haplotypes of caged *Caligus rogercresseyi* in the austral south of Chile: the result of a long term serial passage experiment? *Aquaculture.* 2016;450:143-153.
50. Peeler EJ. Costs and benefits of freedom from shrimp diseases in the European Union. *J Invertebr Pathol.* 2012;110(2):188-195.
51. Thitamadee S, Prachumwat A, Srisala J, et al. Review of current disease threats for cultivated penaeid shrimp in Asia. *Aquaculture.* 2016;452:69-87.
52. Cock J, Salazar M, Rye M. Strategies for managing diseases in non-native shrimp populations. *Rev Aquac.* 2017;9(3):211-226.
53. Alvarez-Lee A, Martínez-Díaz SF, Gutiérrez-Rivera JN, Lanz-Mendoza H. Induction of innate immune response in whiteleg shrimp (*Litopenaeus vannamei*) embryos. *Dev Comp Immunol.* 2020;105:103577.
54. Chang YH, Kumar R, Ng TH, Wang HC. What vaccination studies tell us about immunological memory within the innate immune system of cultured shrimp and crayfish. *Dev Comp Immunol.* 2018;80:53-66.
55. Phuoc LH, Hu B, Wille M, et al. Priming the immune system of Penaeid shrimp by bacterial HSP70 (DnaK). *J Fish Dis.* 2016;39(5):555-564.
56. Jeney G. *Fish Diseases: Prevention and Control Strategies.* Elsevier Science; 2017.
57. Shinn AP, Pratoomyot J, Bron JE, Paladini G, Brooker EE, Brooker AJ. Economic costs of protistan and metazoan parasites to global mariculture. *Parasitology.* 2015;142(1):196-270.
58. Gjedrem T, Robinson N, Rye M. The importance of selective breeding in aquaculture to meet future demands for animal protein: a review. *Aquaculture.* 2012;350-353:117-129.
59. Robinson NA, Gjedrem T, Quillet E. Chapter 2—Improvement of disease resistance by genetic methods. In: Jeney G, ed. *Fish Diseases: Prevention and Control Strategies.* Academic Press; 2017:21-50.
60. Gharbi K, Matthews L, Bron J, Roberts R, Tinch A, Stear M. The control of sea lice in Atlantic salmon by selective breeding. *J Roy Soc Interface.* 2015;12(110):0574.
61. Gjerde B, Ødegård J, Thorland I. Estimates of genetic variation in the susceptibility of Atlantic salmon (*Salmo salar*) to the salmon louse *Lepeophtheirus salmonis*. *Aquaculture.* 2011;314(1-4):66-72.
62. Kolstad K, Heuch PA, Gjerde B, Gjedrem T, Salte R. Genetic variation in resistance of Atlantic salmon (*Salmo salar*) to the salmon louse *Lepeophtheirus salmonis*. *Aquaculture.* 2005;247(1-4):145-151.
63. Mustafa A, MacKinnon BM. Genetic variation in susceptibility of Atlantic salmon to the sea louse *Caligus elongatus* Nordmann, 1832. *Can J Zool-Rev Can Zool.* 1999;77(8):1332-1335.
64. Ødegård J, Moen T, Santi N, Korsvoll SA, Kjøglum S, Meuwissen THE. Genomic prediction in an admixed population of Atlantic salmon (*Salmo salar*). *Front Genet.* 2014;5:402.
65. Robledo D, Gutierrez AP, Barria A, Lhorente JP, Houston RD, Yanez JM. Discovery and functional annotation of quantitative trait loci affecting resistance to sea lice in Atlantic Salmon. *Front Genet.* 2019;10:10.
66. Tsai HY, Hamilton A, Tinch AE, et al. Genomic prediction of host resistance to sea lice in farmed Atlantic salmon populations. *Genet Sel Evol.* 2016;48:11.
67. Gitterle T, Gjerde B, Cock J, et al. Optimization of experimental infection protocols for the estimation of genetic parameters of resistance to White spot syndrome virus (WSSV) in *Penaeus (Litopenaeus) vannamei*. *Aquaculture.* 2006;261(2):501-509.
68. Gitterle T, Ødegård J, Gjerde B, Rye M, Salte R. Genetic parameters and accuracy of selection for resistance to White spot syndrome virus (WSSV) in *Penaeus (Litopenaeus) vannamei* using different statistical models. *Aquaculture.* 2006;251(2/4):210-218.
69. Gitterle T, Rye M, Salte R, et al. Genetic (co)variation in harvest body weight and survival in *Penaeus (Litopenaeus) vannamei* under standard commercial conditions. *Aquaculture.* 2005;243(1/4):83-92.
70. Trang TT, Hung NH, Ninh NH, Knibb W, Nguyen H. Genetic variation in disease resistance against White spot syndrome virus (WSSV) in *Litopenaeus vannamei*. *Front Genet.* 2019;10:264.
71. Trang TT, Hung NH, Ninh NH, Nguyena NH. Selection for improved white spot syndrome virus resistance increased larval survival and growth rate of Pacific Whiteleg shrimp, *Litopenaeus vannamei*. *J Invertebr Pathol.* 2019;166:107219.
72. Fast MD, Ross NW, Mustafa A, et al. Susceptibility of rainbow trout *Oncorhynchus mykiss*, Atlantic salmon *Salmo salar* and coho salmon *Oncorhynchus kisutch* to experimental infection with sea lice *Lepeophtheirus salmonis*. *Dis Aquat Organ.* 2002;52(1):57-68.
73. MacKinnon BM. Host factors important in sea lice infections. *ICES J Mar Sci.* 1998;55(2):188-192.
74. Jones SRM, Fast MD, Johnson SC, Groman DB. Differential rejection of salmon lice by pink and chum salmon: disease consequences and expression of proinflammatory genes. *Dis Aquat Organ.* 2007;75(3):229-238.
75. Gjerde B, Saltkjelvik B. Susceptibility of Atlantic salmon and rainbow trout to the salmon lice *Lepeophtheirus salmonis*. *Aquaculture.* 2009;291(1-2):31-34.
76. Gjøen HM, Refstie T, Ulla O, Gjerde B. Genetic correlations between survival of Atlantic salmon in challenge and field tests. *Aquaculture.* 1997;158(3-4):277-288.
77. Gallaga-Maldonado EP, Montaldo HH, Castillo-Juarez H, et al. Crossbreeding effects for White spot disease resistance in challenge tests and field pond performance in Pacific white shrimp *Litopenaeus vannamei* involving susceptible and resistance lines. *Aquaculture.* 2020;516:734527.
78. Lillehammer M, Boison SA, Norris A, Løvoll M, Bakke H, Gjerde B. Genetic parameters of resistance to amoebic gill disease in two Norwegian Atlantic salmon populations. *Aquaculture.* 2019;508:83-89.
79. Chase-Topping ME, Pooley C, Moghadam HK, et al. Impact of vaccination and selective breeding on the transmission of infectious salmon anemia virus. *Aquaculture.* 2021;535:736365.
80. Houston RD, Haley CS, Hamilton A, et al. Major quantitative trait loci affect resistance to infectious pancreatic necrosis in Atlantic salmon (*Salmo salar*). *Genetics.* 2008;178(2):1109-1115.
81. Moen T, Baranski M, Sonesson A, Kjøglum S. Confirmation and fine-mapping of a major QTL for resistance to infectious pancreatic necrosis in Atlantic salmon (*Salmo salar*): population-level associations between markers and trait. *BMC Genomics.* 2009;10(1):368.
82. Moen T, Torgersen J, Santi N, et al. Epithelial cadherin determines resistance to infectious pancreatic necrosis virus in Atlantic salmon. *Genetics.* 2015;200(4):1313-1326.
83. Meuwissen T, Hayes B, Goddard M. Prediction of total genetic value using genome-wide dense marker maps. *Genetics.* 2001;157:1819-1829.
84. Meuwissen T, Hayes B, Goddard M. Genomic selection: a paradigm shift in animal breeding. *Anim Front.* 2016;6(1):6-14.
85. Jinek M, Chylinski K, Fonfara I, Hauer M, Doudna JA, Charpentier E. A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity. *Science.* 2012;337(6096):816-821.
86. Thompson KD. Chapter 1—immunology: improvement of innate and adaptive immunity. In: Jeney G, ed. *Fish Diseases: Prevention and Control Strategies.* Academic Press; 2017.
87. Robledo D, Gutierrez AP, Barria A, Yanez JM, Houston RD. Gene expression response to sea lice in Atlantic Salmon skin: RNA

- sequencing comparison between resistant and susceptible animals. *Front Genet.* 2018;9:10.
88. Holm HJ, Wadsworth S, Bjelland AK, Krasnov A, Evensen O, Skugor S. Dietary phytochemicals modulate skin gene expression profiles and result in reduced lice counts after experimental infection in Atlantic salmon. *Parasit Vectors.* 2016;9:14.
 89. Krasnov A, Breiland MSW, Hatlen B, Afanasyev S, Skugor S. Sexual maturation and administration of 17 beta-estradiol and testosterone induce complex gene expression changes in skin and increase resistance of Atlantic salmon to ectoparasite salmon louse. *Gen Comp Endocrinol.* 2015;212:34-43.
 90. Krasnov A, Skugor S, Todorovic M, Glover KA, Nilsen F. Gene expression in Atlantic salmon skin in response to infection with the parasitic copepod *Lepeophtheirus salmonis*, cortisol implant, and their combination. *BMC Genomics.* 2012;13:130.
 91. Skugor S, Breiland MSW, Hatlen B, Krasnov A. Estrogen and testosterone differentially modulate skin transcriptional responses and protect against salmon louse infection in Atlantic salmon. *Fish Shellfish Immunol.* 2013;34(6):1677-1678.
 92. Skugor S, Glover KA, Nilsen F, Krasnov A. Local and systemic gene expression responses of Atlantic salmon (*Salmo salar* L.) to infection with the salmon louse (*Lepeophtheirus salmonis*). *BMC Genomics.* 2008;9:498.
 93. Tadiso TM, Krasnov A, Skugor S, Afanasyev S, Hordvik I, Nilsen F. Gene expression analyses of immune responses in Atlantic salmon during early stages of infection by salmon louse (*Lepeophtheirus salmonis*) revealed bi-phasic responses coinciding with the copepod-chalimus transition. *Genomics.* 2011;12:141.
 94. Johnson SC, Albright LJ. Comparative susceptibility and histopathology of the response of naive Atlantic, Chinook and coho salmon to experimental-infection with *Lepeophtheirus salmonis* (Copepoda, Caligidae). *Dis Aquat Organ.* 1992;14(3):179-193.
 95. Sutherland BJB, Koczka KW, Yasuie M, et al. Comparative transcriptomics of Atlantic *Salmo salar*, chum *Oncorhynchus keta* and pink salmon *O. gorbuscha* during infections with salmon lice *Lepeophtheirus salmonis*. *BMC Genomics.* 2014;15:17.
 96. Umasuthan N, Xue X, Caballero-Solares A, et al. Transcriptomic profiling in fins of Atlantic Salmon parasitized with sea lice: evidence for an early imbalance between Chalimus-induced immunomodulation and the host's defense response. *Int J Mol Sci.* 2020;21(7):2417.
 97. Braden LM, Michaud D, Groman DB, Byrne P, Purcell SL, Fast MD. Rejection of salmon lice driven in part by chitin-sensing is not impacted by size at smoltification or time at sea in coho salmon. *Sci Rep.* in press.
 98. Fast MD, Braden LM. Salmon-lice interaction and immunological consequences for the host. In: Treasurer J, Bricknell IR, Bron JE, eds. *Sea Lice Biology and Control*; 2022, Nottingham UK: 5M Books.
 99. Fast MD. Fish immune responses to parasitic copepod (namely sea lice) infection. *Dev Comp Immunol.* 2014;43(2):300-312.
 100. Fast MD, Burka JF, Johnson SC, Ross NW. Enzymes released from *Lepeophtheirus salmonis* in response to mucus from different salmonids. *J Parasitol.* 2003;89(1):7-13.
 101. Fast MD, Johnson SC, Eddy TD, Pinto D, Ross NW. *Lepeophtheirus salmonis* secretory/excretory products and their effects on Atlantic salmon immune gene regulation. *Parasite Immunol.* 2007;29(4):179-189.
 102. Fast MD, Muise DM, Easy RE, Ross NW, Johnson SC. The effects of *Lepeophtheirus salmonis* infections on the stress response and immunological status of Atlantic salmon (*Salmo salar*). *Fish Shellfish Immunol.* 2006;21(3):228-241.
 103. Fast MD, Ross NW, Muise DM, Johnson SC. Differential gene expression in Atlantic salmon infected with *Lepeophtheirus salmonis*. *J Aquat Anim Health.* 2006;18(2):116-127.
 104. Fast MD, Sims DE, Burka JF, Mustafa A, Ross NW. Skin morphology and humoral non-specific defence parameters of mucus and plasma in rainbow trout, coho and Atlantic salmon. *Comp Biochem Physiol A-Mol Integr Physiol.* 2002;132(3):645-657.
 105. Wagner GN, Fast MD, Johnson SC. Physiology and immunology of *Lepeophtheirus salmonis* infections of salmonids. *Trends Parasitol.* 2008;24(4):176-183.
 106. Braden LM, Monaghan SJ, Fast MD. Salmon immunological defence and interplay with the modulatory capabilities of its ectoparasite *Lepeophtheirus salmonis*. *Parasite Immunol.* 2020;42(8):e12731.
 107. Øvergård AC, Hamre LA, Harasimczuk E, Dalvin S, Nilsen F, Grotmol S. Exocrine glands of *Lepeophtheirus salmonis* (Copepoda: Caligidae): distribution, developmental appearance, and site of secretion. *J Morphol.* 2016;277(12):1616-1630.
 108. Valenzuela-Munoz V, Boltana S, Gallardo-Escarate C. Uncovering iron regulation with species-specific transcriptome patterns in Atlantic and coho salmon during a *Caligus rogercresseyi* infestation. *J Fish Dis.* 2017;40(9):1169-1184.
 109. Lewis DL, Barker DE, McKinley RS. Modulation of cellular innate immunity by *Lepeophtheirus salmonis* secretory products. *Fish Shellfish Immunol.* 2014;38(1):175-183.
 110. Hamilton S, McLean K, Monaghan SJ, et al. Characterisation of proteins in excretory/secretory products collected from salmon lice, *Lepeophtheirus salmonis*. *Parasites Vectors.* 2018;11:294.
 111. Nunez-Acuna G, Vera-Bizama F, Boltana S, et al. In-feed additives modulate ionotropic receptor genes from the sea louse *Caligus rogercresseyi*: a comparative analysis in two host salmonid species. *Aquaculture.* 2016;451:99-105.
 112. Vera-Bizama F, Valenzuela-Munoz V, Goncalves AT, et al. Transcription expression of immune-related genes from *Caligus rogercresseyi* evidences host-dependent patterns on Atlantic and coho salmon. *Fish Shellfish Immunol.* 2015;47(2):725-731.
 113. Braden LM, Sutherland BJB, Koop BF, Jones SRM. Enhanced transcriptomic responses in the Pacific salmon louse *Lepeophtheirus salmonis* oncorhynchii to the non-native Atlantic Salmon *Salmo salar* suggests increased parasite fitness. *Genomics.* 2017;18:110.
 114. Huang ZJ, Kang ST, Leu JH, Chen LL. Endocytic pathway is indicated for white spot syndrome virus (WSSV) entry in shrimp. *Fish Shellfish Immunol.* 2013;35(3):707-715.
 115. Zheng SC, Xu JY, Liu HP. Cellular entry of white spot syndrome virus and antiviral immunity mediated by cellular receptors in crustaceans. *Fish Shellfish Immunol.* 2019;93:580-588.
 116. Li CZ, Wang S, He JG. The two NF-kappa B pathways regulating bacterial and WSSV infection of shrimp. *Front Immunol.* 2019;10:1785. doi:10.3389/fimmu.2019.01785
 117. Li M, Ma CX, Zhu P, et al. A new crustin is involved in the innate immune response of shrimp *Litopenaeus vannamei*. *Fish Shellfish Immunol.* 2019;94:398-406.
 118. Ren Q, Huang X, Cui YL, Sun JJ, Wang W, Zhang XB. Two White spot syndrome virus microRNAs target the dorsal gene to promote virus infection in *Marsupenaeus japonicus* shrimp. *J Virol.* 2017;91(8):e02261-e02216.
 119. Wang PH, Gu ZH, Wan DH, et al. The shrimp NF-kappa B pathway is activated by White spot syndrome virus (WSSV) 449 to facilitate the expression of WSSV069 (ie1), WSSV303 and WSSV371. *PLoS One.* 2011;6(9):e24773.
 120. Sun JJ, Lan JF, Zhao XF, Vasta GR, Wang JX. Binding of a C-type lectin's coiled-coil domain to the Domeless receptor directly activates the JAK/STAT pathway in the shrimp immune response to bacterial infection. *PLoS Pathog.* 2017;13(9):e1006626.
 121. Liu WJ, Chang YS, Wang AHJ, Kou GH, Lo CF. White spot syndrome virus annexes a shrimp STAT to enhance expression of the immediate-early gene ie1. *J Virol.* 2007;81(3):1461-1471.

122. Clarke TE, Clem RJ. Insect defenses against virus infection: the role of apoptosis. *Int Rev Immunol*. 2003;22(5-6):401-424.
123. Flegel TW. Update on viral accommodation, a model for host-viral interaction in shrimp and other arthropods. *Dev Comp Immunol*. 2007;31(3):217-231.
124. Granja CB, Aranguren LF, Vidal OM, Aragon L, Salazar M. Does hyperthermia increase apoptosis in white spot syndrome virus (WSSV)-infected *Litopenaeus vannamei*? *Dis Aquat Organ*. 2003;54(1):73-78.
125. Wongprasert K, Khanobdee K, Glunukarn SS, Meeratana P, Withyachumnarnkul B. Time-course and levels of apoptosis in various tissues of black tiger shrimp *Penaeus monodon* infected with white-spot syndrome virus. *Dis Aquat Organ*. 2003;55(1):3-10.
126. Leu JH, Lin SJ, Huang JY, Chen TC, Lo CF. A model for apoptotic interaction between white spot syndrome virus and shrimp. *Fish Shellfish Immunol*. 2013;34(4):1011-1017.
127. Lulijwa R, Alfaro AC, Young T. Metabolomics in salmonid aquaculture research: applications and future perspectives. *Rev Aquac*. 2022;14(2):547-577.
128. Qin R, Mahal LK. The host glycomic response to pathogens. *Curr Opin Struct Biol*. 2021;68:149-156.
129. Chen IT, Aoki T, Huang YT, et al. White spot syndrome virus induces metabolic changes resembling the Warburg effect in shrimp hemocytes in the early stage of infection. *J Virol*. 2011;85(24):12919-12928.
130. Kumar R, Huang JY, Ng YS, Chen CY, Wang HC. The regulation of shrimp metabolism by the white spot syndrome virus (WSSV). *Rev Aquac*. 2022;14:1150-1169.
131. Su MA, Huang YT, Chen IT, et al. An invertebrate Warburg effect: a shrimp virus achieves successful replication by altering the host metabolome via the PI3K-Akt-mTOR pathway. *PLoS Pathog*. 2014;10(6):e1004196, e1004196.
132. Miranda-Cruz MM, Poom-Llamas JJ, Godoy-Lugo JA, et al. Silencing of HIF-1 in WSSV-infected white shrimp: effect on viral load and antioxidant enzymes. *Comp Biochem Physiol C Toxicol Pharmacol*. 2018;213:19-26.
133. Amar EC, Faisan JP. Induction of immunity and resistance to White spot syndrome virus (WSSV) in shrimp *Penaeus monodon* (Fabricius) by synthetic Oligodeoxynucleotide and bacterial DNA. *Philipp Agric Sci*. 2012;95(3):267-277.
134. Kojour MAM, Baliarsingh S, Jang HA, et al. Current knowledge of immune priming in invertebrates, emphasizing studies on *Tenebrio molitor*. *Dev Comp Immunol*. 2022;127:104284.
135. Lafont M, Vergnes A, Vidal-Dupiol J, et al. A sustained immune response supports long-term antiviral immune priming in the Pacific oyster, *Crassostrea gigas*. *MBio*. 2020;11(2):e02777-e02719.
136. Devine GJ, Ingvarsdottir A, Mordue W, et al. Salmon lice, *Lepeophtheirus salmonis*, exhibit specific chemotactic responses to semiochemicals originating from the salmonid, *Salmo salar*. *J Chem Ecol*. 2000;26(8):1833-1847.
137. Brooker AJ, Shinn AP, Souissi S, Bron JE. Role of kairomones in host location of the pennellid copepod parasite, *Lernaecocera branchialis* (L. 1767). *Parasitology*. 2013;140(6):756-770.
138. Pino-Marambio J, Mordue AJ, Birkett M, et al. Behavioural studies of host, non-host and mate location by the sea louse, *Caligus rogercresseyi* Boxshall & Bravo, 2000 (Copepoda: Caligidae). *Aquaculture*. 2007;271(1-4):70-76.
139. Fields DM, Weissburg MJ, Browman H. Chemoreception in the salmon louse *Lepeophtheirus salmonis*: an electrophysiology approach. *Dis Aquat Organ*. 2007;78(2):161-168.
140. Bailey RJE, Birkett MA, Ingvarsdottir A, et al. The role of semiochemicals in host location and non-host avoidance by salmon louse (*Lepeophtheirus salmonis*) copepodids. *Can J Fish Aquat Sci*. 2006;63(2):448-456.
141. Mordue Luntz AJ, Birkett MA. A review of host finding behaviour in the parasitic sea louse, *Lepeophtheirus salmonis* (Caligidae: Copepoda). *J Fish Dis*. 2009;32(1):3-13.
142. Komisarczuk AZ, Grotmol S, Nilsen F. Ionotropic receptors signal host recognition in the salmon louse (*Lepeophtheirus salmonis*, Copepoda). *PLoS One*. 2017;12(6):e0178812.
143. Nunez-Acuna G, Gallardo-Escarate C, Skiftesvik AB, Fields DM, Browman HI. Silencing of ionotropic receptor 25a decreases chemosensory activity in the salmon louse *Lepeophtheirus salmonis* during the infective stage. *Gene*. 2019;697:35-39.
144. Boxshall GA. Infections with parasitic copepods in north-sea marine fishes. *J Mar Biol Assoc UK*. 1974;54(2):355-372.
145. Ingvarsdottir A, Birkett MA, Duce I, et al. Semiochemical strategies for sea louse control: host location cues. *Pest Manag Sci*. 2002;58(6):537-545.
146. Nunez-Acuna G, Gallardo-Escarate C, Fields DM, et al. The Atlantic salmon (*Salmo salar*) antimicrobial peptide cathelicidin-2 is a molecular host-associated cue for the salmon louse (*Lepeophtheirus salmonis*). *Sci Rep*. 2018;8:13738.
147. Lazado CC. The 1st international symposium on mucosal health in aquaculture – MHA2019. Tissue Barriers. *Tissue Barriers*; 2020;8(2):1712177.
148. Bohbot JD, Fu L, Le TC, Chauhan KR, Cantrell CL, Dickens JC. Multiple activities of insect repellents on odorant receptors in mosquitoes. *Med Vet Entomol*. 2011;25(4):436-444.
149. Mands V, Kline DL, Blackwell A. Culicoides midge trap enhancement with animal odour baits in Scotland. *Med Vet Entomol*. 2004;18(4):336-342.
150. Torres-Estrada JL, Rodriguez MH. Physico-chemical signals involved in host localization and induction of disease vector mosquito bites. *Salud Publica De Mexico*. 2003;45(6):497-505.
151. Difford GF, Haugen JE, Aslam ML, et al. Variation in volatile organic compounds in Atlantic salmon mucus is associated with resistance to salmon lice infection. *Sci Rep*. 2022;12(1):4839.
152. Yamada T, Sugiura H, Mimura H, Kamiya K, Osaki T, Takeuchi S. Highly sensitive VOC detectors using insect olfactory receptors reconstituted into lipid bilayers. *Sci Adv*. 2021;7(3):eabd2013.
153. Ozaki K, Ohnishi Y, Iida A, et al. Functional SNPs in the lymphotoxin-alpha gene that are associated with susceptibility to myocardial infarction. *Nat Genet*. 2002;32(4):650-654.
154. Pavelin J, Jin YH, Gratacap RL, et al. The nedd-8 activating enzyme gene underlies genetic resistance to infectious pancreatic necrosis virus in Atlantic salmon. *Genomics*. 2021;113(6):3842-3850.
155. Hölper JE, Grey F, Baillie JK, et al. A genome-wide CRISPR/Cas9 screen reveals the requirement of host sphingomyelin synthase 1 for infection with pseudorabies virus mutant gD(-)pass. *Viruses*. 2021;13(8):1574.
156. Mortazavi A, Williams BA, McCue K, Schaeffer L, Wold B. Mapping and quantifying mammalian transcriptomes by RNA-Seq. *Nat Methods*. 2008;5(7):621-628.
157. Nagalakshmi U, Wang Z, Waern K, et al. The transcriptional landscape of the yeast genome defined by RNA sequencing. *Science*. 2008;320(5881):1344-1349.
158. Anonymous. Method of the year 2013. *Nat Methods*. 2014;11(1):1.
159. Aldridge S, Teichmann SA. Single cell transcriptomics comes of age. *Nat Commun*. 2020;11(1):4307.
160. Kulkarni A, Anderson AG, Merullo DP, Konopka G. Beyond bulk: a review of single cell transcriptomics methodologies and applications. *Curr Opin Biotechnol*. 2019;58:129-136.
161. Slyper M, Porter CBM, Ashenberg O, et al. A single-cell and single-nucleus RNA-Seq toolbox for fresh and frozen human tumors. *Nat Med*. 2020;26(5):792.

162. Asp M, Bergenstrahle J, Lundberg J. Spatially resolved transcriptomes-next generation tools for tissue exploration. *Bioessays*. 2020;42(10):e1900221.
163. Rao A, Barkley D, Franca GS, Yanai I. Exploring tissue architecture using spatial transcriptomics. *Nature*. 2021;596(7871):211-220.
164. Jaitin DA, Weiner A, Yofe I, et al. Dissecting immune circuits by linking CRISPR-pooled screens with single-cell RNA-Seq. *Cell*. 2016;167(7):1883-1896.e1815.
165. Wang M, Yu G, Resson HW. Integrative analysis of proteomic, Glycomic, and Metabolomic data for biomarker discovery. *IEEE J Biomed Health Inform*. 2016;20(5):1225-1231.
166. Jean Beltran P, Mathias R, Cristea I. A portrait of the human organellar proteome in space and time during cytomegalovirus infection. *Cell Syst*. 2016;3:361-373.e6.
167. Lieberman-Aiden E, van Berkum NL, Williams L, et al. Comprehensive mapping of long-range interactions reveals folding principles of the human genome. *Science*. 2009;326(5950):289-293.
168. Fullwood MJ, Liu MH, Pan YF, et al. An oestrogen-receptor- α -bound human chromatin interactome. *Nature*. 2009;462(7269):58-64.
169. Buenrostro JD, Giresi PG, Zaba LC, Chang HY, Greenleaf WJ. Transposition of native chromatin for fast and sensitive epigenomic profiling of open chromatin, DNA-binding proteins and nucleosome position. *Nat Methods*. 2013;10(12):1213-1218.
170. Buenrostro JD, Wu B, Litzenburger UM, et al. Single-cell chromatin accessibility reveals principles of regulatory variation. *Nature*. 2015;523(7561):486-490.
171. Johnson DS, Mortazavi A, Myers RM, Wold B. Genome-wide mapping of in vivo protein-DNA interactions. *Science*. 2007;316(5830):1497-1502.
172. Stirzaker C, Taberlay PC, Statham AL, Clark SJ. Mining cancer methylomes: prospects and challenges. *Trends Genet*. 2014;30(2):75-84.
173. Lister R, Pelizzola M, Downen RH, et al. Human DNA methylomes at base resolution show widespread epigenomic differences. *Nature*. 2009;462(7271):315-322.
174. Meissner A, Gnirke A, Bell GW, Ramsahoye B, Lander ES, Jaenisch R. Reduced representation bisulfite sequencing for comparative high-resolution DNA methylation analysis. *Nucleic Acids Res*. 2005;33(18):5868-5877.
175. Subramanian I, Verma S, Kumar S, Jere A, Anamika K. Multi-omics data integration, interpretation, and its application. *Bioinform Biol Insights*. 2020;14:1177932219899051.
176. Hao Y, Hao S, Andersen-Nissen E, et al. Integrated analysis of multimodal single-cell data. *Cell*. 2021;184(13):3573-3587.e3529.
177. Waiho K, Afiqah-Aleng N, Iryani MTM, Fazhan H. Protein-protein interaction network: an emerging tool for understanding fish disease in aquaculture. *Rev Aquac*. 2021;13(1):156-177.
178. Gervais O, Barria A, Papadopoulou A, et al. Exploring genetic resistance to infectious salmon anaemia virus in Atlantic salmon by genome-wide association and RNA sequencing. *BMC Genomics*. 2021;22(1):345.
179. Dussault FM, Ang KP, Elliott JAK, et al. Mapping quantitative trait loci for infectious salmon anaemia resistance in a north American strain of Atlantic salmon. *Aquacult Res*. 2020;51(1):80-90.
180. Aslam ML, Robledo D, Krasnov A, et al. Quantitative trait loci and genes associated with salmonid alphavirus load in Atlantic salmon: implications for pancreas disease resistance and tolerance. *Sci Rep*. 2020;10(1):10393.
181. Hillestad B, Makvandi-Nejad S, Krasnov A, Moghadam HK. Identification of genetic loci associated with higher resistance to pancreas disease (PD) in Atlantic salmon (*Salmo salar* L.). *BMC Genomics*. 2020;21(1):388.
182. Hillestad B, Kristjansson OH, Makvandi-Nejad S, Moghadam HK. Genome-wide association study confirms previous findings of major loci affecting resistance to *Piscine myocarditis virus* in Atlantic Salmon (*Salmo salar* L.). *Genes*. 2020;11(6):608.
183. Hillestad B, Moghadam HK. Genome-wide association study of piscine myocarditis virus (PMCV) resistance in Atlantic Salmon (*Salmo salar*). *J Hered*. 2019;110(6):720-726.
184. Boison S, Ding J, Leder E, et al. QTLs associated with resistance to cardiomyopathy syndrome in Atlantic salmon. *J Hered*. 2019;110(6):727-737.
185. Boison SA, Gjerde B, Hillestad B, Makvandi-Nejad S, Moghadam HK. Genomic and transcriptomic analysis of amoebic gill disease resistance in Atlantic Salmon (*Salmo salar* L.). *Front Genet*. 2019;10:68.
186. Aslam ML, Boison SA, Lillehammer M, Norris A, Gjerde B. Genome-wide association mapping and accuracy of predictions for amoebic gill disease in Atlantic salmon (*Salmo salar*). *Sci Rep*. 2020;10(1):6435.
187. Robledo D, Matika O, Hamilton A, Houston RD. Genome-wide association and genomic selection for resistance to amoebic gill disease in Atlantic Salmon. *G3-Genes Genomes Genet*. 2018;8(4):1195-1203.
188. Holborn MK, Rochus CM, Ang KP, et al. Family-based genome wide association analysis for salmon lice (*Lepeophtheirus salmonis*) resistance in north American Atlantic salmon using a 50 K SNP array. *Aquaculture*. 2019;511:734215.
189. Rochus CM, Holborn MK, Ang KP, et al. Genome-wide association analysis of salmon lice (*Lepeophtheirus salmonis*) resistance in a north American Atlantic salmon population. *Aquacult Res*. 2018;49(3):1329-1338.
190. Houston RD, Davey JW, Bishop SC, et al. Characterisation of QTL-linked and genome-wide restriction site-associated DNA (RAD) markers in farmed Atlantic salmon. *BMC Genomics*. 2012;13:244.
191. Houston RD, Gheys A, Hamilton A, et al. Detection and confirmation of a major QTL affecting resistance to infectious pancreatic necrosis (IPN) in Atlantic salmon (*Salmo salar*). *Dev Biol*. 2008;132:199-204.
192. Moraleda CP, Robledo D, Gutierrez AP, del-Pozo J, Yanez JM, Houston RD. Investigating mechanisms underlying genetic resistance to Salmon Rickettsial syndrome in Atlantic salmon using RNA sequencing. *BMC Genomics*. 2021;22(1):156.
193. Barria A, Christensen KA, Yoshida GM, et al. Genomic predictions and genome-wide association study of resistance against *Piscirickettsia salmonis* in Coho Salmon (*Oncorhynchus kisutch*) Using ddRAD sequencing. *G3-Genes Genomes Genet*. 2018;8(4):1183-1194.
194. Jiang YL, Yu MH, Dong CJ, et al. Genomic features of common carp that are relevant for resistance against *Aeromonas hydrophila* infection. *Aquaculture*. 2022;547:737512.
195. Jia Z, Chen L, Ge Y, et al. Genetic mapping of koi herpesvirus resistance (KHVR) in Mirror carp (*Cyprinus carpio*) revealed genes and molecular mechanisms of disease resistance. *Aquaculture*. 2020;519:734850.
196. Paliokostas C, Robledo D, Vesely T, et al. Mapping and sequencing of a significant quantitative trait locus affecting resistance to koi herpesvirus in common carp. *G3 (Bethesda, Md)*. 2018;8(11):3507-3513.
197. Tadmor-Levi R, Hulata G, David L. Multiple interacting QTLs affect disease challenge survival in common carp (*Cyprinus carpio*). *Heredity*. 2019;123(5):565-578.
198. Robinson N, Baranski M, Das Mahapatra K, et al. A linkage map of transcribed single nucleotide polymorphisms in rohu (*Labeo rohita*) and QTL associated with resistance to *Aeromonas hydrophila*. *BMC Genomics*. 2014;15:541.
199. Marana MH, Karami AM, Ødegård J, et al. Whole-genome association study searching for QTL for *Aeromonas salmonicida* resistance in rainbow trout. *Sci Rep*. 2021;11(1):17857.

200. Karami AM, Ødegård J, Marana MH, et al. A major QTL for resistance to *Vibrio anguillarum* in rainbow trout. *Front Genet.* 2020;11:607558.
201. Jaafar R, Ødegård J, Mathiessen H, et al. Quantitative trait loci (QTL) associated with resistance of rainbow trout *Oncorhynchus mykiss* against the parasitic ciliate *Ichthyophthirius multifiliis*. *J Fish Dis.* 2020;43(12):1591-1602.
202. Barria A, Marin-Nahuelpi R, Caceres P, et al. Single-step genome-wide association study for resistance to *Piscirickettsia salmonis* in rainbow trout (*Oncorhynchus mykiss*). *G3-Genes Genomes Genet.* 2019;9(11):3833-3841.
203. Rodriguez FH, Flores-Mara R, Yoshida GM, et al. Genome-wide association analysis for resistance to infectious pancreatic necrosis virus identifies candidate genes involved in viral replication and immune response in rainbow trout (*Oncorhynchus mykiss*). *G3-Genes Genomes Genet.* 2019;9(9):2897-2904.
204. Silva RMO, Evenhuis JP, Vallejo RL, et al. Whole-genome mapping of quantitative trait loci and accuracy of genomic predictions for resistance to columnaris disease in two rainbow trout breeding populations. *Genet Sel Evol.* 2019;51(1):42.
205. Liu SX, Vallejo RL, Evenhuis JP, et al. Retrospective evaluation of marker-assisted selection for resistance to bacterial cold water disease in three generations of a commercial rainbow trout breeding population. *Front Genet.* 2018;9:286.
206. Vallejo RL, Liu SX, Gao GT, et al. Similar genetic architecture with shared and unique quantitative trait loci for bacterial cold water disease resistance in two rainbow trout breeding populations. *Front Genet.* 2017;8:156.
207. Liu S, Vallejo RL, Palti Y, et al. Identification of single nucleotide polymorphism markers associated with bacterial cold water disease resistance and spleen size in rainbow trout. *Front Genet.* 2015;6:298.
208. Frasin C, Brard-Fudulea S, D'Ambrosio J, et al. Rainbow trout resistance to bacterial cold water disease: two new quantitative trait loci identified after a natural disease outbreak on a French farm. *Anim Genet.* 2019;50(3):293-297.
209. Moen T, Ødegård J, Baranski M, Lien S, Santi N. A QTL for resistance to infectious pancreatic necrosis in rainbow trout. *Aquaculture.* 2017;472:161-162.
210. Verrier ER, Dorson M, Mauger S, et al. Resistance to a rhabdovirus (VHSV) in rainbow trout: identification of a major QTL related to innate mechanisms. *PLoS One.* 2013;8(2):e55302.
211. Barria A, Trinh TQ, Mahmuddin M, et al. A major quantitative trait locus affecting resistance to tilapia lake virus in farmed Nile tilapia (*Oreochromis niloticus*). *Heredity.* 2021;127(3):334-343.
212. Sukhavachana S, Tongyoo P, Massault C, McMillan N, Leungnaruemitchai A, Poompuang S. Genome-wide association study and genomic prediction for resistance against *Streptococcus agalactiae* in hybrid red tilapia (*Oreochromis* spp.). *Aquaculture.* 2020;525:735297.
213. Griot R, Allal F, Phocas F, et al. Genome-wide association studies for resistance to viral nervous necrosis in three populations of European sea bass (*Dicentrarchus labrax*) using a novel 57k SNP array Dlab-Chip. *Aquaculture.* 2021;530:735930.
214. Paliokostas C, Cariou S, Bestin A, et al. Genome-wide association and genomic prediction of resistance to viral nervous necrosis in European sea bass (*Dicentrarchus labrax*) using RAD sequencing. *Genet Sel Evol.* 2018;50:30.
215. Chatziplis D, Oikonomou S, Loukovitis D, et al. QTL for stress and disease resistance in European sea bass, *Dicentrarchus labrax* L. *Animals (Basel).* 2020;10(9):1668.
216. Liu L, Liu P, Huang SQ, et al. Genome-wide association study identifies loci associated with resistance to viral nervous necrosis disease in Asian seabass. *Marine Biotechnol.* 2017;19(3):255-265.
217. Liu P, Wang L, Wong S-M, Yue GH. Fine mapping QTL for resistance to VNN disease using a high-density linkage map in Asian seabass. *Sci Rep.* 2016;6(1):32122.
218. Yang M, Wang Q, Chen JP, Wang YX, Zhang Y, Qin QW. Identification of candidate SNPs and genes associated with anti-RGNNV using GWAS in the red-spotted grouper, *Epinephelus akaara*. *Fish Shellfish Immunol.* 2021;112:31-37.
219. Uchino T, Tabata J, Yoshida K, et al. Novel *Benedenia* disease resistance QTLs in five F1 families of yellowtail (*Seriola quinqueradiata*). *Aquaculture.* 2020;529:735622.
220. Nakamoto M, Takeuchi Y, Akita K, et al. A novel C-type lectin gene is a strong candidate gene for *Benedenia* disease resistance in Japanese yellowtail, *Seriola quinqueradiata*. *Dev Comp Immunol.* 2017;76:361-369.
221. Ozaki A, Yoshida K, Fuji K, et al. Quantitative trait loci (QTL) associated with resistance to a monogenean parasite (*Benedenia seriola*) in yellowtail (*Seriola quinqueradiata*) through genome wide analysis. *PLoS One.* 2013;8(6):e64987.
222. Mastrochirico VA, Borges CHS, Freitas MV, et al. Development of a SNP linkage map and genome-wide association study for resistance to *Aeromonas hydrophila* in pacu (*Piaractus mesopotamicus*). *BMC Genomics.* 2020;21(1):672.
223. Wang WW, Tan SX, Luo J, et al. GWAS analysis indicated importance of NF- κ B signaling pathway in host resistance against motile *Aeromonas* septicemic disease in catfish. *Marine Biotechnol.* 2019;21(3):335-347.
224. Shi HT, Zhou T, Wang XZ, et al. Genome-wide association analysis of intra-specific QTL associated with the resistance for enteric septicemia of catfish. *Mol Genet Genomics.* 2018;293(6):1365-1378.
225. Tan SX, Zhou T, Wang WW, et al. GWAS analysis using interspecific backcross progenies reveals superior blue catfish alleles responsible for strong resistance against enteric septicemia of catfish. *Mol Genet Genomics.* 2018;293(5):1107-1120.
226. Zhou T, Liu SK, Geng X, et al. GWAS analysis of QTL for enteric septicemia of catfish and their involved genes suggest evolutionary conservation of a molecular mechanism of disease resistance. *Mol Genet Genomics.* 2017;292(1):231-242.
227. Geng X, Sha J, Liu S, et al. A genome-wide association study in catfish reveals the presence of functional hubs of related genes within QTLs for columnaris disease resistance. *BMC Genomics.* 2015;16:196.
228. Kong SN, Ke QZ, Chen L, et al. Constructing a high-density genetic linkage map for large yellow croaker (*Larimichthys crocea*) and mapping resistance trait against ciliate parasite *Cryptocaryon irritans*. *Marine Biotechnol.* 2019;21(2):262-275.
229. Wan L, Wang WJ, Liu GJ, et al. A genome-wide association study of resistance to *Pseudomonas plecoglossicida* infection in the large yellow croaker (*Larimichthys crocea*). *Aquac Int.* 2019;27(5):1195-1208.
230. Aslam ML, Carraro R, Bestin A, et al. Genetics of resistance to photobacteriosis in gilthead sea bream (*Sparus aurata*) using 2b-RAD sequencing. *BMC Genetics.* 2018;19:43.
231. Aslam ML, Carraro R, Sonesson AK, et al. Genetic variation, GWAS and accuracy of prediction for host resistance to *Sparicotyle chryso-phrii* in farmed Gilthead Sea bream (*Sparus aurata*). *Front Genet.* 2020;11:594770.
232. Shao C, Niu Y, Rastas P, et al. Genome-wide SNP identification for the construction of a high-resolution genetic map of Japanese flounder (*Paralichthys olivaceus*): applications to QTL mapping of *Vibrio anguillarum* disease resistance and comparative genomic analysis. *DNA Res.* 2015;22(2):161-170.
233. Rodriguez-Ramilo ST, Toro MA, Bouza C, et al. QTL detection for *Aeromonas salmonicida* resistance related traits in turbot (*Scophthalmus maximus*). *BMC Genomics.* 2011;12:541.

234. Saura M, Carabaño MJ, Fernández A, et al. Disentangling genetic variation for resistance and endurance to Scuticociliatosis in turbot using pedigree and genomic information. *Front Genet.* 2019;10:539.
235. Rodríguez-Ramilo ST, Fernandez J, Toro MA, et al. Uncovering QTL for resistance and survival time to *Philasterides dicentrarchi* in turbot (*Scophthalmus maximus*). *Anim Genet.* 2013;44(2):149-157.
236. Sawayama E, Tanizawa S, Kitamura SI, et al. Identification of quantitative trait loci for resistance to RSIVD in Red Sea bream (*Pagrus major*). *Mar Biotechnol (NY).* 2017;19(6):601-613.
237. Robinson NA, Gopikrishna G, Baranski M, et al. QTL for white spot syndrome virus resistance and the sex-determining locus in the Indian black tiger shrimp (*Penaeus monodon*). *BMC Genomics.* 2014; 15:731.
238. Yang B, Zhai SY, Zhang FQ, et al. Genome-wide association study toward efficient selection breeding of resistance to *Vibrio alginolyticus* in Pacific oyster, *Crassostrea gigas*. *Aquaculture.* 2022;548: 737592.
239. Gutierrez AP, Bean TP, Hooper C, et al. A genome-wide association study for host resistance to *Ostreid herpesvirus* in Pacific oysters (*Crassostrea gigas*). *G3-Genes Genomes Genet.* 2018;8(4):1273-1280.
240. Harrang E, Heurtebise S, Faury N, Robert M, Arzul I, Lapegue S. Can survival of European flat oysters following experimental infection with *Bonamia ostreae* be predicted using QTLs? *Aquaculture.* 2015; 448:521-530.
241. Farhat S, Tanguy A, Pales Espinosa E, et al. Identification of variants associated with hard clam, *Mercenaria mercenaria*, resistance to quahog parasite unknown disease. *Genomics.* 2020;112(6):4887-4896.
242. Hayes B, Goddard ME. The distribution of the effects of genes affecting quantitative traits in livestock. *Genet Sel Evol.* 2001;33(3): 209-229.
243. Kjetsa MH, Ødegård J, Meuwissen THE. Accuracy of genomic prediction of host resistance to salmon lice in Atlantic salmon (*Salmo salar*) using imputed high-density genotypes. *Aquaculture.* 2020;526: 735415.
244. Piazzon MC, Mladineo I, Dirks RP, Yebra-Pimentel ES, Hrabar J, Sitja-Bobadilla A. *Ceratomyxa oestroides* infection in European sea bass: revealing a long misunderstood relationship. *Front Immunol.* 2021;12:645607.
245. Lama R, Pereiro P, Valenzuela-Munoz V, et al. RNA-Seq analysis of European sea bass (*Dicentrarchus labrax* L.) infected with nodavirus reveals powerful modulation of the stress response. *Vet Res.* 2020; 51(1):64.
246. Chaves-Pozo E, Bandin I, Oliveira JG, et al. European sea bass brain DLB-1 cell line is susceptible to nodavirus: a transcriptomic study. *Fish Shellfish Immunol.* 2019;86:14-24.
247. Chaves-Pozo E, Valero Y, Esteve-Codina A, et al. Innate cell-mediated cytotoxic activity of European sea bass leucocytes against nodavirus-infected cells: a functional and RNA-seq study. *Sci Rep.* 2017;7:15396.
248. Sarropoulou E, Galindo-Villegas J, Garcia-Alcazar A, Kasapidis P, Mulero V. Characterization of European sea bass transcripts by RNA SEQ after oral vaccine against *V. anguillarum*. *Marine Biotechnol.* 2012;14(5):634-642.
249. Zhao C, Fu MJ, Wang CY, Jiao ZY, Qiu LH. RNA-Seq analysis of immune-relevant genes in *Lateolabrax japonicus* during *Vibrio anguillarum* infection. *Fish Shellfish Immunol.* 2016;52:57-64.
250. Xiang L-X, He D, Dong W-r, Zhang Y-W, Shao J-Z. Deep sequencing-based transcriptome profiling analysis of bacteria-challenged *Lateolabrax japonicus* reveals insight into the immune-relevant genes in marine fish. *BMC Genomics.* 2010;11(1):472.
251. Gao EB, Chen GF. Micropterus salmoides rhabdovirus (MSRV) infection induced apoptosis and activated interferon signaling pathway in largemouth bass skin cells. *Fish Shellfish Immunol.* 2018;76:161-166.
252. Tran NT, Gao ZX, Zhao HH, et al. Transcriptome analysis and microsatellite discovery in the blunt snout bream (*Megalobrama amblycephala*) after challenge with *Aeromonas hydrophila*. *Fish Shellfish Immunol.* 2015;45(1):72-82.
253. Piazzon MC, Mladineo I, Naya-Catala F, et al. Acting locally—affecting globally: RNA sequencing of gilthead sea bream with a mild *Sparicotyle chrysophrii* infection reveals effects on apoptosis, immune and hypoxia related genes. *BMC Genomics.* 2019;20:200.
254. Song LF, Dong XZ, Hu GB. Transcriptome analysis of red sea bream (*Pagrus major*) head kidney and spleen infected by *Vibrio anguillarum*. *Aquac Rep.* 2021;21:100789.
255. Peruzza L, Pascoli F, Dalla Rovere G, et al. Transcriptome analysis reveals a complex response to the RGNNV/SJNNV reassortant nervous necrosis virus strain in sea bream larvae. *Fish Shellfish Immunol.* 2021;114:282-292.
256. Zhang XS, She YB, Xu XY, et al. Transcriptome analysis and histopathology of black carp (*Mylopharyngodon piceus*) spleen infected by *Aeromonas hydrophila*. *Fish Shellfish Immunol.* 2018;83:330-340.
257. Das BK, Chakraborty HJ, Rout AK, Behera BK. De novo whole transcriptome profiling of *Edwardsiella tarda* isolated from infected fish (*Labeo catla*). *Gene.* 2019;701:152-160.
258. Jiang YL, Feng SS, Zhang SH, et al. Transcriptome signatures in common carp spleen in response to *Aeromonas hydrophila* infection. *Fish Shellfish Immunol.* 2016;57:41-48.
259. Verma DK, Peruzza L, Trusch F, et al. Transcriptome analysis reveals immune pathways underlying resistance in the common carp *Cyprinus carpio* against the oomycete *Aphanomyces invadans*. *Genomics.* 2021;113(1):944-956.
260. Tadmor-Levi R, Doron-Faigenboim A, Marcos-Hadad E, et al. Different transcriptional response between susceptible and resistant common carp (*Cyprinus carpio*) fish hints on the mechanism of CyHV-3 disease resistance. *BMC Genomics.* 2019;20(1):1019.
261. Neave MJ, Sunarto A, McColl KA. Transcriptomic analysis of common carp anterior kidney during *Cyprinid herpesvirus 3* infection: immunoglobulin repertoire and homologue functional divergence. *Sci Rep.* 2017;7:41531.
262. Reed AN, Putman T, Sullivan C, Jin L. Application of a nanoflare probe specific to a latency associated transcript for isolation of KHV latently infected cells. *Virus Res.* 2015;208:129-135.
263. Yuan JF, Yang Y, Nie HH, et al. Transcriptome analysis of epithelioma papulosum cyprini cells after SVCV infection. *BMC Genomics.* 2014;15:935.
264. Zhang YS, Zhang X, Liang Z, et al. Expression pattern and regulatory network of gibel carp (*Carassius gibelio*) miRNAs and their putative target genes in response to CyHV-2 infection. *Aquaculture.* 2020; 523:735178.
265. Zhang TT, Yuan R, Zhang X, et al. De novo RNA-seq assembly and differential transcriptome analysis of *Carassius auratus gibelio* after *Cyprinid herpesvirus 2* infection. *Aquaculture.* 2020;519:734923.
266. Zhao YL, Weng MQ, Zhang QQ, Li AH, Zhang JY. Transcriptomics analysis of the infected tissue of gibel carp (*Carassius auratus gibelio*) with liver myxobolosis infers the underlying defense mechanisms from the perspective of immune-metabolic interactions. *Aquaculture.* 2021;542:736867.
267. Zhao YL, Liu XH, Sato H, Zhang QQ, Li AH, Zhang JY. RNA-seq analysis of local tissue of *Carassius auratus gibelio* with pharyngeal myxobolosis: insights into the pharyngeal mucosal immune response in a fish-parasite dialogue. *Fish Shellfish Immunol.* 2019;94:99-112.
268. Dang YF, Xu XY, Shen YB, et al. Transcriptome analysis of the innate immunity-related complement system in spleen tissue of *Ctenopharyngodon idella* infected with *Aeromonas hydrophila*. *PLoS One.* 2016; 11(7):e0157413.
269. Song XH, Hu XL, Sun BY, et al. A transcriptome analysis focusing on inflammation-related genes of grass carp intestines following infection with *Aeromonas hydrophila*. *Sci Rep.* 2017;7:40777.
270. Yao QC, Zhang MD, Zu SP, et al. Integrated mRNA and microRNA transcriptome sequencing characterizes sequence variants and

- mRNA-microRNA regulatory networks in grass carp fibroblasts infected with virulent and attenuated GCRV. *Marine Biotechnol.* 2021;23(2):342-355.
271. He LB, Zhu DH, Liang XY, et al. Multi-omics sequencing provides insights into age-dependent susceptibility of grass carp (*Ctenopharyngodon idellus*) to Reovirus. *Front Immunol.* 2021;12:694965.
272. He LB, Zhang AD, Xiong L, et al. Deep circular RNA sequencing provides insights into the mechanism underlying grass carp Reovirus infection. *Int J Mol Sci.* 2017;18(9):1977.
273. He LB, Zhang AD, Pei YY, et al. Differences in responses of grass carp to different types of grass carp reovirus (GCRV) and the mechanism of hemorrhage revealed by transcriptome sequencing. *BMC Genomics.* 2017;18:452.
274. Shi MJ, Huang R, Du FK, et al. RNA-seq profiles from grass carp tissues after reovirus (GCRV) infection based on singular and modular enrichment analyses. *Mol Immunol.* 2014;61(1):44-53.
275. Shen MH, Jiang ZY, Zhang K, et al. Transcriptome analysis of grass carp (*Ctenopharyngodon idella*) and Holland's spinibarbel (*Spinibarbus hollandi*) infected with *Ichthyophthirius multifiliis*. *Fish Shellfish Immunol.* 2022;121:305-315.
276. Li JN, Zhao YT, Cao SL, Wang H, Zhang JJ. Integrated transcriptomic and proteomic analyses of grass carp intestines after vaccination with a double-targeted DNA vaccine of *Vibrio mimicus*. *Fish Shellfish Immunol.* 2020;98:641-652.
277. Pradhan PK, Verma DK, Peruzza L, et al. Molecular insights into the mechanisms of susceptibility of *Labeo rohita* against oomycete *Aphanomyces invadans*. *Sci Rep.* 2020;10(1):19531.
278. Robinson N, Sahoo PK, Baranski M, et al. Expressed sequences and polymorphisms in rohu carp (*Labeo rohita*, Hamilton) revealed by mRNA-seq. *Marine Biotechnol.* 2012;14(5):620-633.
279. Hu MH, Qu XC, Pan LS, et al. Effects of toxic *Microcystis aeruginosa* on the silver carp *Hypophthalmichthys molitrix* revealed by hepatic RNA-seq and miRNA-seq. *Sci Rep.* 2017;7:10456.
280. Li ZZ, Wang XM, Chen CX, Gao JW, Lv AJ. Transcriptome profiles in the spleen of African catfish (*Clarias gariepinus*) challenged with *Aeromonas veronii*. *Fish Shellfish Immunol.* 2019;86:858-867.
281. Dawar FU, Hu XQ, Zhao LJ, et al. Transcriptomic analysis reveals differentially expressed genes and a unique apoptosis pathway in channel catfish ovary cells after infection with the channel catfish virus. *Fish Shellfish Immunol.* 2017;71:58-68.
282. Tan SX, Wang WW, Zhong XX, et al. Increased alternative splicing as a host response to *Edwardsiella ictaluri* infection in catfish. *Marine Biotechnol.* 2018;20(6):729-738.
283. Li C, Zhang Y, Wang RJ, et al. RNA-seq analysis of mucosal immune responses reveals signatures of intestinal barrier disruption and pathogen entry following *Edwardsiella ictaluri* infection in channel catfish, *Ictalurus punctatus*. *Fish Shellfish Immunol.* 2012;32(5):816-827.
284. Yuan ZH, Liu SK, Yao J, Zeng QF, Tan SX, Liu ZJ. Expression of Bcl-2 genes in channel catfish after bacterial infection and hypoxia stress. *Dev Comp Immunol.* 2016;65:79-90.
285. Liu HX, Xu HZ, Shangguan XY, Wang LX, Liu XQ. Identification, annotation of mucin genes in channel catfish (*Ictalurus punctatus*) and their expression after bacterial infections revealed by RNA-Seq analysis. *Aquacult Res.* 2020;51(5):2020-2028.
286. Li ZX, Yao J, Xie YJ, Geng X, Liu ZJ. Phosphoinositide 3-kinase family in channel catfish and their regulated expression after bacterial infection. *Fish Shellfish Immunol.* 2016;49:364-373.
287. Mu WJ, Yao J, Zhang JR, et al. Expression of tumor suppressor genes in channel catfish after bacterial infections. *Dev Comp Immunol.* 2015;48(1):171-177.
288. Song L, Zhang JR, Li C, et al. Genome-wide identification of Hsp40 genes in channel catfish and their regulated expression after bacterial infection. *PLoS One.* 2014;9(12):e115752.
289. Wang XZ, Liu SK, Yang YJ, Fu Q, Abebe A, Liu ZJ. Identification of NF-kappa B related genes in channel catfish and their expression profiles in mucosal tissues after columnaris bacterial infection. *Dev Comp Immunol.* 2017;70:27-38.
290. Lange MD, Abernathy J, Farmer BD. Evaluation of a recombinant *Flavobacterium columnare* DnaK protein vaccine as a means of protection against Columnaris disease in channel catfish (*Ictalurus punctatus*). *Front Immunol.* 2019;10:1175.
291. Lange MD, Farmer BD, Abernathy J. Catfish mucus alters the *Flavobacterium columnare* transcriptome. *FEMS Microbiol Lett.* 2018;365(22):fny244.
292. Tan SX, Wang WW, Tian CX, et al. Post-transcriptional regulation through alternative splicing after infection with *Flavobacterium columnare* in channel catfish (*Ictalurus punctatus*). *Fish Shellfish Immunol.* 2019;91:188-193.
293. Liu LS, Li C, Su BF, Beck BH, Peatman E. Short-term feed deprivation alters immune status of surface mucosa in channel catfish (*Ictalurus punctatus*). *PLoS One.* 2013;8(9):e74581.
294. Beck BH, Farmer BD, Straus DL, Li C, Peatman E. Putative roles for a rhamnose binding lectin in *Flavobacterium columnare* pathogenesis in channel catfish *Ictalurus punctatus*. *Fish Shellfish Immunol.* 2012;33(4):1008-1015.
295. Sun FY, Peatman E, Li C, et al. Transcriptomic signatures of attachment, NF-kappa B suppression and IFN stimulation in the catfish gill following columnaris bacterial infection. *Dev Comp Immunol.* 2012;38(1):169-180.
296. Wang RJ, Sun LY, Bao LS, et al. Bulk segregant RNA-seq reveals expression and positional candidate genes and allele-specific expression for disease resistance against enteric septicemia of catfish. *BMC Genomics.* 2013;14:929.
297. Sun LY, Liu SK, Bao LS, Li Y, Feng JB, Liu ZJ. *Claudin* multigene family in channel catfish and their expression profiles in response to bacterial infection and hypoxia as revealed by meta-analysis of RNA-Seq datasets. *Comp Biochem Physiol D-Genomics Proteomics.* 2015;13:60-69.
298. Yang YB, Zhu X, Zhang HX, Chen YH, Song Y, Ai XH. Dual RNA-Seq of trunk kidneys extracted from channel catfish infected with *Yersinia ruckeri* reveals novel insights into host-pathogen interactions. *Front Immunol.* 2021;12:775708.
299. Zhu CK, Pan ZJ, Chang GL, et al. Transcriptomic insights into immune responses to ulcerative syndrome in & *Pseudobagrus ussuriensis*. *Aquaculture.* 2021;537:736504.
300. Zhou X, Zhang GR, Ji W, et al. The dynamic immune response of yellow catfish (*Pelteobagrus fulvidraco*) infected with *Edwardsiella ictaluri* presenting the inflammation process. *Front Immunol.* 2021;12:625928.
301. Maekawa S, Wang PC, Chen SC. Differential expression of immune-related genes in head kidney and spleen of cobia (*Rachycentron canadum*) having *Streptococcus dysgalactiae* infection. *Fish Shellfish Immunol.* 2019;92:842-850.
302. Zhang YM, Lu LX, Li CW, Shao GM, Chen XH. Transcriptome analysis revealed multiple immune processes and energy metabolism pathways involved in the defense response of the large yellow croaker *Larimichthys crocea* against *Pseudomonas plecoglossicida*. *Comp Biochem Physiol D-Genomics Proteomics.* 2021;40:100886.
303. Wang LY, Sun YJ, Zhao LM, et al. Dual RNA-seq uncovers the immune response of *Larimichthys crocea* to the secY gene of *Pseudomonas plecoglossicida* from the perspective of host-pathogen interactions. *Fish Shellfish Immunol.* 2019;93:949-957.
304. Tang Y, Xin G, Zhao LM, et al. Novel insights into host-pathogen interactions of large yellow croakers (*Larimichthys crocea*) and pathogenic bacterium *Pseudomonas plecoglossicida* using time-resolved dual RNA-seq of infected spleens. *Zool Res.* 2020;41(3):314-327.
305. Tang Y, Sun YJ, Zhao LM, et al. Mechanistic insight into the roles of *Pseudomonas plecoglossicida* clpV gene in host-pathogen interactions

- with *Larimichthys crocea* by dual RNA-seq. *Fish Shellfish Immunol.* 2019;93:344-353.
306. Xiong F, Xiong J, Wu YF, Cao L, Huang WS, Chang MX. Time-resolved RNA-seq provided a new understanding of intestinal immune response of European eel (*Anguilla Anguilla*) following infection with *Aeromonas hydrophila*. *Fish Shellfish Immunol.* 2020;105:297-309.
 307. Zheng ZY, Yang JX, Ge JQ, et al. Development and characterization of a continuous cell line (EL) from the liver of European eel *Anguilla anguilla*. *Cell Biol Int.* 2020;44(3):808-820.
 308. Bracamonte SE, Johnston PR, Monaghan MT, Knopf K. Gene expression response to a nematode parasite in novel and native eel hosts. *Ecol Evol.* 2019;9(23):13069-13084.
 309. Xiu YJ, Jiang GP, Zhou S, et al. Identification of potential immune-related circRNA-miRNA-mRNA regulatory network in intestine of *Paralichthys olivaceus* during *Edwardsiella tarda* infection. *Front Genet.* 2019;10:731.
 310. Li WR, Guan XL, Sun B, Sun L. A novel microRNA of Japanese flounder regulates antimicrobial immunity involving a bacteria-binding CSF3. *Front Immunol.* 2021;12:723401.
 311. Wang HX, Tang XQ, Sheng XZ, Xing J, Chi H, Zhan WB. Transcriptome analysis reveals temperature-dependent early immune response in flounder (*Paralichthys olivaceus*) after *Hirame novirhabdovirus* (HIRRV) infection. *Fish Shellfish Immunol.* 2020;107:367-378.
 312. Kim KI, Lee UH, Cho M, Jung SH, Min EY, Park JW. Transcriptome analysis based on RNA-seq of common innate immune responses of flounder cells to IHNV, VHSV, and HIRRV. *PLoS One.* 2020;15(9):e0239925.
 313. Xing J, Jiang XY, Xu HS, et al. Local immune responses to VAA DNA vaccine against *Listonella anguillarum* in flounder (*Paralichthys olivaceus*). *Mol Immunol.* 2021;134:141-149.
 314. Wu RH, Sheng XZ, Tang XQ, Xing J, Zhan WB. Transcriptome analysis of flounder (*Paralichthys olivaceus*) gill in response to lymphocystis disease virus (LCDV) infection: novel insights into fish defense mechanisms. *Int J Mol Sci.* 2018;19(1):160.
 315. Wu Q, Ning XH, Sun L. Megalocytivirus induces complicated fish immune response at multiple RNA levels involving mRNA, miRNA, and circRNA. *Int J Mol Sci.* 2021;22(6):3156.
 316. Hwang JY, Kwon MG, Seo JS, et al. Differentially expressed genes after viral haemorrhagic septicaemia virus infection in olive flounder (*Paralichthys olivaceus*). *Vet Microbiol.* 2016;193:72-82.
 317. Hwang JY, Lee UH, Heo MJ, et al. Naturally occurring substitution in one amino acid in VHSV phosphoprotein enhances viral virulence in flounder. *PLoS Pathog.* 2021;17(1):e1009213.
 318. Hwang JY, Lee UH, Heo MJ, et al. RNA-seq transcriptome analysis in flounder cells to compare innate immune responses to low- and high-virulence viral hemorrhagic septicemia virus. *Arch Virol.* 2021;166(1):191-206.
 319. Jeong JM, Jeswin J, Bae JS, Park CI. Differential expression of olive flounder (*Paralichthys olivaceus*) transcriptome during viral hemorrhagic septicemia virus (VHSV) infection at warmer and colder temperature. *Data Brief.* 2018;19:2023-2028.
 320. Hwang JY, Markkandan K, Han K, et al. Temperature-dependent immune response of olive flounder (*Paralichthys olivaceus*) infected with viral hemorrhagic septicemia virus (VHSV). *Genes Genomics.* 2018;40(3):315-320.
 321. Hwang JY, Kwon MG, Jung SH, et al. RNA-Seq transcriptome analysis of the olive flounder (*Paralichthys olivaceus*) kidney response to vaccination with heat-inactivated viral hemorrhagic septicemia virus. *Fish Shellfish Immunol.* 2017;62:221-226.
 322. Lu JF, Xu D, Jiang YS, et al. Integrated analysis of mRNA and viral miRNAs in the kidney of *Carassius auratus gibelio* response to cyprinid herpesvirus 2. *Sci Rep.* 2017;7:13787.
 323. Diao J, Yu XQ, Wang XL, et al. Full-length transcriptome sequencing combined with RNA-seq analysis revealed the immune response of fat greenling (*Hexagrammos otakii*) to *Vibrio harveyi* in early infection. *Microb Pathog.* 2020;149:104527.
 324. Jazamuddin FM, Aizat WM, Goh HH, Low CF, Baharum SN. Alternative RNA extraction method in *Vibrio vulnificus* infected Brown-marbled grouper, *Epinephelus Fuscoguttatus*. *Trop Life Sci Res.* 2019;30(2):201-209.
 325. Zheng GL, Zhou Q, Li KM, et al. Transcriptome analysis of Giant grouper (*Epinephelus lanceolatus*) kidney and spleen in response to spotted knifejaw iridovirus (SKIV) infection. *Aquacult Res.* 2021;52(5):1954-1964.
 326. Tso CH, Lu MW. Transcriptome profiling analysis of grouper during nervous necrosis virus persistent infection. *Fish Shellfish Immunol.* 2018;76:224-232.
 327. Wang LN, Tian YS, Cheng ML, et al. Transcriptome comparative analysis of immune tissues from asymptomatic and diseased *Epinephelus moara* naturally infected with nervous necrosis virus. *Fish Shellfish Immunol.* 2019;93:99-107.
 328. Liu CC, Ho LP, Yang CH, Kao TY, Chou HY, Pai TW. Comparison of grouper infection with two different iridoviruses using transcriptome sequencing and multiple reference species selection. *Fish Shellfish Immunol.* 2017;71:264-274.
 329. Ge H, Lin KB, Zhou C, et al. A multi-omic analysis of orange-spotted grouper larvae infected with nervous necrosis virus identifies increased adhesion molecules and collagen synthesis in the persistent state. *Fish Shellfish Immunol.* 2020;98:595-604.
 330. Chen WJ, Yi LZ, Feng SS, et al. Characterization of microRNAs in orange-spotted grouper (*Epinephelus coioides*) fin cells upon red-spotted grouper nervous necrosis virus infection. *Fish Shellfish Immunol.* 2017;63:228-236.
 331. Jiao JP, Zhao LM, Huang LX, et al. The contributions of fliG gene to the pathogenicity of *Pseudomonas plecoglossicida* and pathogen-host interactions with *Epinephelus coioides*. *Fish Shellfish Immunol.* 2021;119:238-248.
 332. Liu ZX, Zhao LM, Huang LX, et al. Integration of RNA-seq and RNAi provides a novel insight into the immune responses of *Epinephelus coioides* to the impB gene of *Pseudomonas plecoglossicida*. *Fish Shellfish Immunol.* 2020;105:135-143.
 333. Tang RQ, Zhao LM, Xu XJ, et al. Dual RNA-Seq uncovers the function of an ABC transporter gene in the host pathogen interaction between *Epinephelus coioides* and *Pseudomonas plecoglossicida*. *Fish Shellfish Immunol.* 2019;92:45-53.
 334. Tang RQ, Luo G, Zhao LM, et al. The effect of a LysR-type transcriptional regulator gene of *Pseudomonas plecoglossicida* on the immune responses of *Epinephelus coioides*. *Fish Shellfish Immunol.* 2019;89:420-427.
 335. Huang LX, Zhao LM, Liu WJ, et al. Dual RNA-Seq unveils *Pseudomonas plecoglossicida* htpG gene functions during host-pathogen interactions with *Epinephelus coioides*. *Front Immunol.* 2019;10:984.
 336. Sun YJ, Zhuang ZX, Wang XR, Huang HB, Fu Q, Yan QP. Dual RNA-seq reveals the effect of the flgM gene of *Pseudomonas plecoglossicida* on the immune response of *Epinephelus coioides*. *Fish Shellfish Immunol.* 2019;87:515-523.
 337. Sun YJ, Luo G, Zhao LM, et al. Integration of RNAi and RNA-seq reveals the immune responses of *Epinephelus coioides* to sigX gene of *Pseudomonas plecoglossicida*. *Front Immunol.* 2018;9:1624.
 338. Wang Q, Peng C, Yang M, et al. Single-cell RNA-seq landscape mid-brain cell responses to red spotted grouper nervous necrosis virus infection. *PLoS Pathog.* 2021;17(6):e1009665.
 339. Chen WJ, Yi LZ, Feng SS, et al. Transcriptomic profiles of striped snakehead fish cells (SSN-1) infected with red-spotted grouper nervous necrosis virus (RGNNV) with an emphasis on apoptosis pathway. *Fish Shellfish Immunol.* 2017;60:346-354.
 340. Xiang YX, Jia P, Liu W, Yi MS, Jia KT. Comparative transcriptome analysis reveals the role of p53 signalling pathway during red-

- spotted grouper nervous necrosis virus infection in *Lateolabrax japonicus* brain cells. *J Fish Dis.* 2019;42(4):585-595.
341. Cai Y, Wang SF, Guo WL, et al. Transcriptome analysis provides insights into the immune responsive pathways and genes in the head kidney of tiger grouper (*Epinephelus fuscoguttatus*) fed with *Spatholobus suberectus*, *Phellodendron amurense*, or *Eclipta prostrata*. *Fish Shellfish Immunol.* 2018;73:100-111.
 342. Legrand T, Wynne JW, Weyrich LS, Oxley APA. Investigating both mucosal immunity and microbiota in response to gut enteritis in yellowtail kingfish. *Microorganisms.* 2020;8(9):1267.
 343. Eggsetol HO, Lunde HS, Ronneseth A, et al. Transcriptome-wide mapping of signaling pathways and early immune responses in lumpfish leukocytes upon in vitro bacterial exposure. *Sci Rep.* 2018;8:5261.
 344. Hu XQ, Fu XZ, Li NQ, et al. Transcriptomic analysis of mandarin fish brain cells infected with infectious spleen and kidney necrosis virus with an emphasis on retinoic acid-inducible gene 1-like receptors and apoptosis pathways. *Fish Shellfish Immunol.* 2015;45(2):619-629.
 345. Hampton LMT, Martyniuk CJ, Venables BJ, Jeffries MKS. Advancing the fathead minnow (*Pimephales promelas*) as a model for immunotoxicity testing: characterization of the renal transcriptome following *Yersinia ruckeri* infection. *Fish Shellfish Immunol.* 2020;103:472-480.
 346. Lin YS, Wang B, Wang NH, Ouyang G, Cao H. Transcriptome analysis of rare minnow (*Gobiocypris rarus*) infected by the grass carp reovirus. *Fish Shellfish Immunol.* 2019;89:337-344.
 347. Mastrochirico VA, Hata ME, Kuradomi RY, et al. Transcriptome profiling of Pacu (*Piaractus mesopotamicus*) challenged with pathogenic *Aeromonas hydrophila*: inference on immune gene response. *Front Genet.* 2020;11:604.
 348. Lacaze E, Gendron AD, Miller JL, et al. Cumulative effects of municipal effluent and parasite infection in yellow perch: a field study using high-throughput RNA-sequencing. *Sci Total Environ.* 2019;665:797-809.
 349. Yu YX, Wang YG, Liao MJ, et al. Comparative analysis of spleen transcriptome of immune response in *Sebastes schlegeli* against *Photobacterium damsela* subsp. *damsela* infection. *Aquacult Res.* 2022;53(1):232-242.
 350. Andresen AMS, Gjoen T. Transcriptome profiling of Atlantic salmon kidney cells (ASIC) after stimulation with poly (I:C) and infection with infectious Salmon anemia virus (ISAV). *Fish Shellfish Immunol.* 2019;91:390.
 351. Cai WL, Kumar S, Navaneethaiyer U, et al. Transcriptome analysis of Atlantic Salmon (*Salmo salar*) skin in response to sea lice and infectious Salmon anemia virus co-infection under different experimental functional diets. *Front Immunol.* 2022;12:787033.
 352. Valenzuela-Miranda D, Boltana S, Cabrejos ME, Yanez JM, Gallardo-Escarate C. High-throughput transcriptome analysis of ISAV-infected Atlantic salmon *Salmo salar* unravels divergent immune responses associated to head-kidney, liver and gills tissues. *Fish Shellfish Immunol.* 2015;45(2):367-377.
 353. Salazar C, Marshall SH. Involvement of selected cellular miRNAs in the in vitro and in vivo infection of infectious salmon anemia virus (ISAV). *Microb Pathog.* 2018;123:353-360.
 354. Valenzuela-Miranda D, Cabrejos ME, Yanez JM, Gallardo-Escarate C. From the viral perspective: infectious salmon anemia virus (ISAV) transcriptome during the infective process in Atlantic salmon (*Salmo salar*). *Marine Genomics.* 2015;20:39-43.
 355. Dettleff P, Moen T, Santi N, Martinez V. Transcriptomic analysis of spleen infected with infectious salmon anemia virus reveals distinct pattern of viral replication on resistant and susceptible Atlantic salmon (*Salmo salar*). *Fish Shellfish Immunol.* 2017;61:187-193.
 356. Salazar C, Galaz M, Ojeda N, Marshall SH. Detection of cellular miRNAs in plasma of *Salmo salar* during an ISAV infection. *Aquac Rep.* 2020;17:100320.
 357. Talbot A, Gargan L, Moran G, et al. Investigation of the transcriptomic response in Atlantic salmon (*Salmo salar*) gill exposed to *Paramoeba perurans* during early onset of disease. *Sci Rep.* 2021;11(1):20682.
 358. Robledo D, Hamilton A, Gutierrez AP, Bron JE, Houston RD. Characterising the mechanisms underlying genetic resistance to amoebic gill disease in Atlantic salmon using RNA sequencing. *BMC Genomics.* 2020;21(1):11.
 359. Botwright NA, Mohamed AR, Slinger J, Lima PC, Wynne JW. Host-parasite interaction of Atlantic salmon (*Salmo salar*) and the Ectoparasite *Neoparamoeba perurans* in amoebic gill disease. *Front Immunol.* 2021;12:672700.
 360. Heggland EI, Dondrup M, Nilsen F, Eichner C. Host gill attachment causes blood-feeding by the salmon louse (*Lepeophtheirus salmonis*) chalimus larvae and alters parasite development and transcriptome. *Parasites Vectors.* 2020;13(1):225.
 361. Valenzuela-Munoz V, Novoa B, Figueras A, Gallardo-Escarate C. Modulation of Atlantic salmon miRNome response to sea louse infestation. *Dev Comp Immunol.* 2017;76:380-391.
 362. Nunez-Acuna G, Marambio JP, Valenzuela T, Wadsworth S, Gallardo-Escarate C. Antimicrobial peptides from salmon *Salmo salar* skin induce frontal filament development and olfactory/cuticle-related genes in the sea louse *Caligus rogercresseyi*. *Aquaculture.* 2016;464:171-177.
 363. Fu Q, Li YQ, Zhang H, et al. Comparative transcriptome analysis of spleen reveals potential regulation of genes and immune pathways following administration of *Aeromonas salmonicida* subsp. *masoucida* vaccine in Atlantic Salmon (*Salmo salar*). *Marine Biotechnol.* 2022;18:1-14.
 364. Ren GM, Xu LM, Zhao JZ, Shao YZ, Lu TY, Zhang QY. Comparative transcriptome analysis of long non coding RNA (lncRNA) in RTG-2 cells infected by infectious hematopoietic necrosis virus. *Fish Shellfish Immunol.* 2022;120:314-324.
 365. Houston RD, Taggart JB, Bishop SC, Bron JE, Bekaert MB. Gene expression profiles defining host resistance to infectious pancreatic necrosis virus in Atlantic salmon fry. *Aquaculture.* 2017;472:110.
 366. Woldemariam NT, Agafonov O, Sindre H, et al. miRNAs predicted to regulate host anti-viral gene pathways in IPNV-challenged Atlantic Salmon fry are affected by viral load, and associated with the major IPN resistance QTL genotypes in late infection. *Front Immunol.* 2020;11:2113.
 367. Mohr PG, Crane MS, Hoad J, et al. Pilchard orthomyxovirus (POMV). I. Characterisation of an emerging virus isolated from pilchards *Sardinops sagax* and Atlantic salmon *Salmo salar*. *Dis Aquat Organ.* 2020;139:35-50.
 368. Valenzuela-Miranda D, Gallardo-Escarate C. Dual RNA-Seq uncovers metabolic amino acids dependency of the intracellular bacterium *Piscirickettsia salmonis* infecting Atlantic Salmon. *Front Microbiol.* 2018;9:2877.
 369. Rozas-Serri M, Pena A, Maldonado L. Transcriptomic profiles of post-smolt Atlantic salmon challenged with *Piscirickettsia salmonis* reveal a strategy to evade the adaptive immune response and modify cell-autonomous immunity. *Dev Comp Immunol.* 2018;81:348-362.
 370. Valenzuela-Miranda D, Gallardo-Escarate C. Novel insights into the response of Atlantic salmon (*Salmo salar*) to *Piscirickettsia salmonis*: interplay of coding genes and lncRNAs during bacterial infection. *Fish Shellfish Immunol.* 2016;59:427-438.
 371. Xu C, Evensen O, Munang'andu HM. De novo transcriptome analysis shows that SAV-3 infection upregulates pattern recognition receptors of the endosomal toll-like and RIG-I-like receptor signaling pathways in macrophage/dendritic like TO-cells. *Viruses.* 2016;8(4):114.
 372. Xu C, Evensen O, Munang'andu HM. De novo assembly and transcriptome analysis of Atlantic salmon macrophage/dendritic-like TO

- cells following type I IFN treatment and salmonid alphavirus sub-type-3 infection. *BMC Genomics*. 2015;16:96.
373. Robinson NA, Krasnov A, Burgerhout E, et al. Response of the Salmon heart transcriptome to pancreas disease: differences between high- and Low-ranking families for resistance. *Sci Rep*. 2020;10(1):868.
374. Ellison AR, Webster TMU, Rodriguez-Barreto D, et al. Comparative transcriptomics reveal conserved impacts of rearing density on immune response of two important aquaculture species. *Fish Shellfish Immunol*. 2020;104:192-201.
375. Polinski MP, Bradshaw JC, Inkpen SM, et al. De novo assembly of sockeye salmon kidney transcriptomes reveal a limited early response to piscine reovirus with or without infectious hematopoietic necrosis virus superinfection. *BMC Genomics*. 2016;17:848.
376. Polinski MP, Bradshaw JC, Rise ML, Johnson SC, Garver KA. Sockeye salmon demonstrate robust yet distinct transcriptomic kidney responses to rhabdovirus (IHNV) exposure and infection. *Fish Shellfish Immunol*. 2019;94:525-538.
377. Domingos JA, Shen XY, Terence C, et al. Scale drop disease virus (SDDV) and Lates calcarifer herpes virus (LCHV) coinfection down-regulate immune-relevant pathways and cause splenic and kidney necrosis in barramundi under commercial farming conditions. *Front Genet*. 2021;12:666897.
378. Yang ZT, Wong SM, Yue GH. Effects of *rrm1* on NNV resistance revealed by RNA-seq and gene editing. *Marine Biotechnol*. 2021;23(6):854-869.
379. Liu P, Wang L, Kwang J, Yue GH, Wong SM. Transcriptome analysis of genes responding to NNV infection in Asian seabass epithelial cells. *Fish Shellfish Immunol*. 2016;54:342-352.
380. Liu C, Ma J, Zhang DF, et al. Immune response and apoptosis-related pathways induced by *Aeromonas schubertii* infection of hybrid snakehead (*Channa maculata* female x *Channa argus* male). *Pathogens*. 2021;10(8):997.
381. Qin XM, Feng SS, Zhang YW, et al. Leader RNA regulates snakehead vesiculovirus replication via interacting with viral nucleoprotein. *RNA Biol*. 2021;18(4):537-546.
382. Sun LD, Babu VS, Qin ZD, et al. Snakehead vesiculovirus (SHVV) infection alters striped snakehead (*Ophicephalus striatus*) cells (SSN-1) glutamine metabolism and apoptosis pathways. *Fish Shellfish Immunol*. 2020;102:36-46.
383. Zhang X, Wang SL, Chen SL, et al. Transcriptome analysis revealed changes of multiple genes involved in immunity in *Cynoglossus semilaevis* during vibrio anguillarum infection. *Fish Shellfish Immunol*. 2015;43(1):209-218.
384. Labella AM, Garcia-Rosado E, Bandin I, et al. Transcriptomic profiles of senegalese sole infected with nervous necrosis virus reassortants presenting different degree of virulence. *Front Immunol*. 2018;9:1626.
385. Barrett DE, Bartholomew JL. A tale of two fish: comparative transcriptomics of resistant and susceptible steelhead following exposure to *Ceratonova shasta* highlights differences in parasite recognition. *PLoS One*. 2021;16(2):e0234837.
386. Zhang QQ, Wang XH, Zhang DF, et al. De novo assembly and analysis of Amur sturgeon (*Acipenser schrenckii*) transcriptome in response to *Mycobacterium marinum* infection to identify putative genes involved in immunity. *J Microbiol Biotechnol*. 2019;29(8):1324-1334.
387. Li SW, Wang D, Cao YS, Zhang Y, Liu HB, Lu TY. Transcriptome profile of Amur sturgeon (*Acipenser schrenckii*) liver provides insights into immune modulation in response to *Yersinia ruckeri* infection. *Aquaculture*. 2018;492:137-146.
388. Li SW, Zhang Y, Cao YS, Wang D, Liu HB, Lu TY. Transcriptome profiles of Amur sturgeon spleen in response to *Yersinia ruckeri* infection. *Fish Shellfish Immunol*. 2017;70:451-460.
389. Ellison AR, Webster TMU, Rey O, et al. Transcriptomic response to parasite infection in Nile tilapia (*Oreochromis niloticus*) depends on rearing density. *BMC Genomics*. 2018;19:723.
390. Maekawa S, Pulpipat T, Wang PC, Chen SC. Transcriptome analysis of immune- and iron-related genes after *Francisella noatunensis* subsp. *orientalis* infection in Nile tilapia (*Oreochromis niloticus*). *Fish Shellfish Immunol*. 2021;111:36-48.
391. Fan BL, Chen FN, Li Y, et al. A comprehensive profile of the tilapia (*Oreochromis niloticus*) circular RNA and circRNA-miRNA network in the pathogenesis of meningoencephalitis of teleosts. *Molecular Omics*. 2019;15(3):233-246.
392. Shen YD, Liang WW, Lin Y, et al. Single molecule real-time sequencing and RNA-seq unravel the role of long non-coding and circular RNA in the regulatory network during Nile tilapia (*Oreochromis niloticus*) infection with *Streptococcus agalactiae*. *Fish Shellfish Immunol*. 2020;104:640-653.
393. Hu QM, Ao QW, Tan Y, Gan X, Luo YJ, Zhu JJ. Genome-wide DNA methylation and RNA analysis reveal potential mechanism of resistance to *Streptococcus agalactiae* in GIFT strain of Nile tilapia (*Oreochromis niloticus*). *J Immunol*. 2020;204(12):3182-3190.
394. Zhang R, Zhang LL, Ye X, et al. Transcriptome profiling and digital gene expression analysis of Nile tilapia (*Oreochromis niloticus*) infected by *Streptococcus agalactiae*. *Mol Biol Rep*. 2013;40(10):5657-5668.
395. Zhang Z, Li YH, Hu MQ, Yu AG. Comparative transcriptome profiling reveals a mechanism of *Streptococcus agalactiae* resistance to florfenicol. *Microb Pathog*. 2020;142:104098.
396. Zhao ZY, Sun CY, Chen LM, Qin JK, Yuan X, Li WS. Inorganic nitrite increases the susceptibility of tilapia (*Oreochromis niloticus*) leucocytes to *Streptococcus agalactiae*. *Fish Shellfish Immunol*. 2020;97:1-11.
397. Wu WD, Li LP, Liu Y, Huang T, Liang WW, Chen M. Multiomics analyses reveal that NOD-like signaling pathway plays an important role against *Streptococcus agalactiae* in the spleen of tilapia. *Fish Shellfish Immunol*. 2019;95:336-348.
398. Liu Y, Li LP, Huang T, Wu WD, Liang WW, Chen M. The interaction between phagocytes and *Streptococcus agalactiae* (GBS) mediated by the activated complement system is the key to GBS inducing acute bacterial meningitis of tilapia. *Animals*. 2019;9(10):818.
399. Zhu JJ, Gan X, Ao QW, et al. Basal polarization of the immune responses to *Streptococcus agalactiae* susceptible and resistant tilapia (*Oreochromis niloticus*). *Fish Shellfish Immunol*. 2018;75:336-345.
400. Zhu JJ, Fu Q, Ao QW, et al. Transcriptomic profiling analysis of tilapia (*Oreochromis niloticus*) following *Streptococcus agalactiae* challenge. *Fish Shellfish Immunol*. 2017;62:202-212.
401. Wang L, Liu P, Wan ZY, et al. RNA-Seq revealed the impairment of immune defence of tilapia against the infection of *Streptococcus agalactiae* with simulated climate warming. *Fish Shellfish Immunol*. 2016;55:679-689.
402. Zhu JJ, Li C, Ao QW, et al. Transcriptomic profiling revealed the signatures of acute immune response in tilapia (*Oreochromis niloticus*) following *Streptococcus iniae* challenge. *Fish Shellfish Immunol*. 2015;46(2):346-353.
403. Sood N, Verma DK, Paria A, et al. Transcriptome analysis of liver eluciates key immune-related pathways in Nile tilapia *Oreochromis niloticus* following infection with tilapia lake virus. *Fish Shellfish Immunol*. 2021;111:208-219.
404. Turnbull OMH, Ortiz-Baez AS, Eden JS, et al. Meta-transcriptomic identification of divergent Amnoonviridae in fish. *Viruses-Basel*. 2020;12(11):1254.
405. Wang YY, Wang Q, Li YY, et al. Integrated analysis of mRNA-miRNA expression in tilapia infected with tilapia lake virus (TiLV) and identifies primarily immuneresponse genes. *Fish Shellfish Immunol*. 2020;99:208-226.

406. Fux R, Arndt D, Langenmayer MC, et al. *Piscine Orthoreovirus 3* is not the causative pathogen of proliferative darkening syndrome (PDS) of Brown trout (*Salmo trutta fario*). *Viruses*. 2019;11(2):112.
407. Sudhagar A, Ertl R, Kumar G, El-Matbouli M. Transcriptome profiling of posterior kidney of brown trout, *Salmo trutta*, during proliferative kidney disease. *Parasites Vectors*. 2019;12(1):569, 569.
408. Kim J, Cho M, Kim KI, Min EY, Lim J, Hong S. Transcriptome profiling in head kidney of rainbow trout (*Oncorhynchus mykiss*) after infection with the low-virulent Nagano genotype of infectious hematopoietic necrosis virus. *Arch Virol*. 2021;166(4):1057-1070.
409. Han YC, Lin CM, Chen TT. RNA-Seq analysis of differentially expressed genes relevant to innate and adaptive immunity in cecropin P1 transgenic rainbow trout (*Oncorhynchus mykiss*). *BMC Genomics*. 2018;19:760.
410. Wang D, Sun SM, Li SW, Lu TY, Shi DF. Transcriptome profiling of immune response to *Yersinia ruckeri* in spleen of rainbow trout (*Oncorhynchus mykiss*). *BMC Genomics*. 2021;22(1):292.
411. Paneru B, Al-Tobasei R, Palti Y, Wiens GD, Salem M. Differential expression of long non-coding RNAs in three genetic lines of rainbow trout in response to infection with *Flavobacterium psychrophilum*. *Sci Rep*. 2016;6:36032.
412. Rivas-Aravena A, Fuentes-Valenzuela M, Escobar-Aguirre S, Gallardo-Escarate C, Molina A, Valdes JA. Transcriptomic response of rainbow trout (*Oncorhynchus mykiss*) skeletal muscle to *Flavobacterium psychrophilum*. *Comp Biochem Physiol D-Genomics Proteomics*. 2019;31:100596.
413. Marancik D, Gao GT, Paneru B, et al. Whole-body transcriptome of selectively bred, resistant-, control-, and susceptible-line rainbow trout following experimental challenge with *Flavobacterium psychrophilum*. *Front Genet*. 2015;5:453.
414. Defo MA, Gendron AD, Head J, et al. Cumulative effects of cadmium and natural stressors (temperature and parasite infection) on molecular and biochemical responses of juvenile rainbow trout. *Aquat Toxicol*. 2019;217:105347.
415. Roh H, Kim N, Lee Y, et al. Dual-organ transcriptomic analysis of rainbow trout infected with *Ichthyophthirius multifiliis* through co-expression and machine learning. *Front Immunol*. 2021;12:677730.
416. Zhang XT, Ding LG, Yu YY, et al. The change of teleost skin commensal microbiota is associated with skin mucosal transcriptomic responses during parasitic infection by *Ichthyophthirius multifiliis*. *Front Immunol*. 2018;9:2972.
417. Syahputra K, Kania PW, Al-Jubury A, Jafaar RM, Dirks RP, Buchmann K. Transcriptomic analysis of immunity in rainbow trout (*Oncorhynchus mykiss*) gills infected by *Ichthyophthirius multifiliis*. *Fish Shellfish Immunol*. 2019;86:486-496.
418. Tapia D, Kuznar J, Farlora R, Yanez JM. Differential transcriptomic response of rainbow trout to infection with two strains of IPNV. *Viruses*. 2022;14(1):21.
419. Zuloaga R, Dettleff P, Bastias-Molina M, et al. RNA-Seq-based analysis of cortisol-induced differential gene expression associated with *Piscirickettsia salmonis* infection in rainbow trout (*Oncorhynchus mykiss*) Myotubes. *Animals*. 2021;11(8):2399.
420. Carrizo V, Valenzuela CA, Aros C, et al. Transcriptomic analysis reveals a *Piscirickettsia salmonis*-induced early inflammatory response in rainbow trout skeletal muscle. *Comp Biochem Physiol Part D Genomics Proteomics*. 2021;39:100859, 100859.
421. Bailey C, von Siebenthal EW, Rehberger K, Segner H. Transcriptomic analysis of the impacts of ethinylestradiol (EE2) and its consequences for proliferative kidney disease outcome in rainbow trout (*Oncorhynchus mykiss*). *Comp Biochem Physiol C Toxicol Pharmacol*. 2019;222:31-48.
422. Hou ZS, Xin YR, Yang XD, et al. Transcriptional profiles of genes related to stress and immune response in rainbow trout (*Oncorhynchus mykiss*) symptomatically or asymptotically infected with *Vibrio anguillarum*. *Front Immunol*. 2021;12:639489.
423. Barrett DE, Estensoro I, Sitja-Bobadilla A, Bartholomew JL. Intestinal transcriptomic and histologic profiling reveals tissue repair mechanisms underlying resistance to the parasite *Ceratomyxa shasta*. *Pathogens*. 2021;10(9):1179.
424. Liu Y, Zhao LY, Yang MJ, et al. Transcriptomic dissection of the horizontally acquired response regulator EsrB reveals its global regulatory roles in the physiological adaptation and activation of T3SS and the cognate effector repertoire in *Edwardsiella piscicida* during infection toward turbot. *Virulence*. 2017;8(7):1355-1377.
425. Ronza P, Robledo D, Bermudez R, et al. RNA-seq analysis of early enteromyxosis in turbot (*Scophthalmus maximus*): new insights into parasite invasion and immune evasion strategies. *Int J Parasit*. 2016;46(8):507-517.
426. Robledo D, Ronza P, Harrison PW, et al. RNA-seq analysis reveals significant transcriptome changes in turbot (*Scophthalmus maximus*) suffering severe enteromyxosis. *BMC Genomics*. 2014;15:1149.
427. Gao CB, Fu Q, Su BF, et al. Transcriptomic profiling revealed the signatures of intestinal barrier alteration and pathogen entry in turbot (*Scophthalmus maximus*) following *Vibrio anguillarum* challenge. *Dev Comp Immunol*. 2016;65:159-168.
428. Li XH, Cui ZX, Liu Y, Song CW, Shi GH. Transcriptome analysis and discovery of genes involved in immune pathways from Hepatopancreas of microbial challenged mitten crab *Eriocheir sinensis*. *PLoS One*. 2013;8(7):e68233.
429. Jin QR, Tian GY, Wu J, Jiang HL, Zhu F. Identification and characterization of hemocyte microRNAs in mud crab *Scylla paramamosain* in response to *Vibrio parahaemolyticus* infection. *Aquaculture*. 2020;524:735288.
430. Lai YY, Jin QR, Zhu F. Differential expression of microRNAs in mud crab *Scylla paramamosain* in response to white spot syndrome virus (WSSV) infection. *Fish Shellfish Immunol*. 2020;105:1-7.
431. Du ZQ, Ren Q, Huang AM, et al. A novel peroxinectin involved in antiviral and antibacterial immunity of mud crab. *Scylla paramamosain Molecular Biology Reports*. 2013;40(12):6873-6881.
432. Guo LF, Zhou M, Chen DD, et al. A new insight to characterize immunomodulation based on hepatopancreatic transcriptome and humoral immune factor analysis of the *Cherax quadricarinatus* infected with *Aeromonas veronii*. *Ecotoxicol Environ Saf*. 2021;219:112347.
433. Nian YY, Chen BK, Wang JJ, et al. Transcriptome analysis of *Procambarus clarkii* infected with infectious hypodermal and hematopoietic necrosis virus. *Fish Shellfish Immunol*. 2020;98:766-772.
434. Liu QN, Huang L, Wang SY, et al. Transcriptome analysis of differentially expressed genes in the red swamp crayfish *Procambarus clarkii* challenged with *Aeromonas hydrophila*. *Fish Shellfish Immunol*. 2021;119:280-288.
435. Liu QN, Tang YY, Zhang SP, et al. Characterization and expression analysis of differentially expressed genes in the red swamp crayfish *Procambarus clarkii* in response to *Vibrio cholerae* challenge. *Aquaculture*. 2022;547:737435.
436. Yang H, Gao XJ, Li X, et al. Comparative transcriptome analysis of red swamp crayfish (*Procambarus clarkii*) hepatopancreas in response to WSSV and *Aeromonas hydrophila* infection. *Fish Shellfish Immunol*. 2018;83:397-405.
437. Dong WQ, Chen YX, Lu WX, Wu B, Qi PZ. Transcriptome analysis of *Mytilus coruscus* hemocytes in response to *Vibrio alginolyticus* infection. *Fish Shellfish Immunol*. 2017;70:560-567.
438. Powell D, Knibb W, Nguyen NH, Elizur A. Transcriptional profiling of Banana shrimp *Fenneropenaeus merguensis* with differing levels of viral load. *Integrative and Comparative Biology*. 2016;56(6):1131-1143.
439. Jaree B, Boonchuen P, Thawonsuan J, Kondo H, Hirono I, Somboonwivat K. Transcriptome profiling reveals the novel immunometabolism-related genes against WSSV infection from *Fenneropenaeus merguensis*. *Fish Shellfish Immunol*. 2022;120:31-44.

440. Soo TCC, Devadas S, Din MSM, Bhassu S. Differential transcriptome analysis of the disease tolerant Madagascar-Malaysia crossbred black tiger shrimp, *Penaeus monodon* hepatopancreas in response to acute hepatopancreatic necrosis disease (AHPND) infection: inference on immune gene response and interaction. *Gut Pathogens*. 2019;11:39.
441. Soonthornchai W, Chaiyapechara S, Klinbunga S, et al. Differentially expressed transcripts in stomach of *Penaeus monodon* in response to AHPND infection. *Dev Comp Immunol*. 2016;65:53-63.
442. He ZH, Chen XY, Zhao JC, et al. Establishment of infection mode and *Penaeus monodon* hemocytes transcriptomics analysis under decapod iridescent virus 1 (DIV1) challenge. *Aquaculture*. 2021;542:736816.
443. Zhao JC, Chen XY, He ZH, et al. Transcriptome analysis provides new insights into host response to Hepatopancreatic necrosis disease in the black Tiger shrimp *Penaeus monodon*. *J Ocean Univ China*. 2021;20(5):1183-1194.
444. Ghani FM, Bhassu S. A new insight to biomarkers related to resistance in survived-white spot syndrome virus challenged giant tiger shrimp, *Penaeus monodon*. *Peerj*. 2019;7:e8107, e8107.
445. Yu CY, Xu WB, Li X, et al. Comparative transcriptome analysis of Chinese grass shrimp (*Palaemonetes sinensis*) hepatopancreas under ectoparasitic isopod (*Tachaea chinensis*) infection. *Fish Shellfish Immunol*. 2021;117:211-219.
446. Chen XW, Wang J, Yue WC, Liu JS, Wang CH. Hepatopancreas transcriptome analysis of Chinese mitten crab (*Eriocheir sinensis*) with white hepatopancreas syndrome. *Fish Shellfish Immunol*. 2017;70:302-307.
447. Thaimuangphol W, Sanoamuang L, Wangkahart E. The immune response of fairy shrimp *Streptocephalus sirindhornae* against bacterial black disease by de novo transcriptome analysis. *Fish Shellfish Immunol*. 2022;121:108-115.
448. Gao XJ, Jiang ZY, Zhang SM, et al. Transcriptome analysis and immune-related genes expression reveals the immune responses of *Macrobrachium rosenbergii* infected by *Enterobacter cloacae*. *Fish Shellfish Immunol*. 2020;101:66-77.
449. Pasookhush P, Hindmarch C, Sithigorngul P, Longyant S, Bendena WG, Chaivisuthangkura P. Transcriptomic analysis of *Macrobrachium rosenbergii* (giant fresh water prawn) post-larvae in response to *M. rosenbergii* nodavirus (MrNV) infection: de novo assembly and functional annotation. *BMC Genomics*. 2019;20(1):762, 762.
450. Rao R, Zhu YB, Alinejad T, et al. RNA-seq analysis of *Macrobrachium rosenbergii* hepatopancreas in response to *Vibrio parahaemolyticus* infection. *Gut Pathogens*. 2015;7:6.
451. Ding ZF, Jin M, Ren Q. Transcriptome analysis of *Macrobrachium rosenbergii* intestines under the white spot syndrome virus and poly (I:C) challenges. *PLoS One*. 2018;13(9):e0204626.
452. Zhu F, Wang Z, Sun BZ. Differential expression of microRNAs in shrimp *Marsupenaeus japonicus* in response to vibrio alginolyticus infection. *Dev Comp Immunol*. 2016;55:76-79.
453. Hui KM, Ren Q, Cao J. Insights into the intestine immune of *Marsupenaeus japonicus* under the white spot syndrome virus challenge using RNA sequencing. *Vet Immunol Immunopathol*. 2019;208:25-33.
454. Zhong SP, Mao Y, Wang J, Liu M, Zhang M, Su YQ. Transcriptome analysis of Kuruma shrimp (*Marsupenaeus japonicus*) hepatopancreas in response to white spot syndrome virus (WSSV) under experimental infection. *Fish Shellfish Immunol*. 2017;70:710-719.
455. Huang TZ, Xu DD, Zhang XB. Characterization of host microRNAs that respond to DNA virus infection in a crustacean. *BMC Genomics*. 2012;13:159.
456. Ren YQ, Li JY, Guo L, et al. Full-length transcriptome and long non-coding RNA profiling of whiteleg shrimp *Penaeus vannamei* hemocytes in response to *Spiroplasma eriocheiris* infection. *Fish Shellfish Immunol*. 2020;106:876-886.
457. Sookruksawong S, Sun FY, Liu ZJ, Tassanakajon A. RNA-Seq analysis reveals genes associated with resistance to Taura syndrome virus (TSV) in the Pacific white shrimp *Litopenaeus vannamei*. *Dev Comp Immunol*. 2013;41(4):523-533.
458. Zeng DG, Chen XL, Xie DX, et al. Transcriptome analysis of Pacific White shrimp (*Litopenaeus vannamei*) Hepatopancreas in response to Taura syndrome virus (TSV) experimental infection. *PLoS One*. 2013;8(2):e57515.
459. Boonchuen P, Jaree P, Somboonviwat K, Somboonviwat K. Regulation of shrimp prophenoloxidase activating system by lva-miR-4850 during bacterial infection. *Sci Rep*. 2021;11(1):3821.
460. Velazquez-Lizarraga AE, Juarez-Morales JL, Racotta IS, et al. Transcriptomic analysis of Pacific white shrimp (*Litopenaeus vannamei*, Boone 1931) in response to acute hepatopancreatic necrosis disease caused by *Vibrio parahaemolyticus*. *PLoS One*. 2019;14(8):e0220993.
461. Boonchuen P, Maralit BA, Jaree P, Tassanakajon A, Somboonviwat K. MicroRNA and mRNA interactions coordinate the immune response in non-lethal heat stressed *Litopenaeus vannamei* against AHPND-causing *Vibrio parahaemolyticus*. *Sci Rep*. 2020;10(1):787.
462. Tinwongger S, Thawonsuwan J, Kondo H, Hirono I. Identification of an anti-lipopolysaccharide factor AV-R isoform (LvALF AV-R) related to Vp_PirAB-like toxin resistance in *Litopenaeus vannamei*. *Fish Shellfish Immunol*. 2019;84:178-188.
463. Maralit BA, Jaree P, Boonchuen P, Tassanakajon A, Somboonviwat K. Differentially expressed genes in hemocytes of *Litopenaeus vannamei* challenged with *Vibrio parahaemolyticus* AHPND (VPAHPND) and VPAHPND toxin. *Fish Shellfish Immunol*. 2018;81:284-296.
464. Qin ZD, Babu VS, Wan QY, et al. Transcriptome analysis of Pacific white shrimp (*Litopenaeus vannamei*) challenged by *Vibrio parahaemolyticus* reveals unique immune-related genes. *Fish Shellfish Immunol*. 2018;77:164-174.
465. Zheng ZH, Wang F, Aweya JJ, et al. Comparative transcriptomic analysis of shrimp hemocytes in response to acute hepatopancreas necrosis disease (AHPND) causing vibrio parahemolyticus infection. *Fish Shellfish Immunol*. 2018;74:10-18.
466. Santos CA, Andrade SCS, Fernandes JMO, Freitas PD. Shedding the light on *Litopenaeus vannamei* Differential muscle and Hepatopancreas immune responses in White spot syndrome virus (WSSV) exposure. *Genes*. 2020;11(7):805.
467. Peruzza L, Shekhar MS, Kumar KV, et al. Temporal changes in transcriptome profile provide insights of White spot syndrome virus infection in *Litopenaeus vannamei*. *Sci Rep*. 2019;9:13509.
468. Santos CA, Andrade SCS, Freitas PD. Identification of SNPs potentially related to immune responses and growth performance in *Litopenaeus vannamei* by RNA-seq analyses. *Peerj*. 2018;6:e5154.
469. Xue SX, Liu YC, Zhang YC, Sun Y, Geng XY, Sun JS. Sequencing and De novo analysis of the Hemocytes transcriptome in *Litopenaeus vannamei* response to White spot syndrome virus infection. *PLoS One*. 2013;8(10):e76718.
470. Chen XH, Zeng DG, Chen XL, et al. Transcriptome analysis of *Litopenaeus vannamei* in response to White spot syndrome virus infection. *PLoS One*. 2013;8(8):e73218.
471. Millard RS, Bickley LK, Bateman KS, et al. Global mRNA and miRNA analysis reveal key processes in the initial response to infection with WSSV in the Pacific Whiteleg shrimp. *Viruses*. 2021;13(6):1140.
472. Peruzza L, Thamizhvanan S, Vimal S, et al. A comparative synthesis of transcriptomic analyses reveals major differences between WSSV-susceptible *Litopenaeus vannamei* and WSSV-refractory *Macrobrachium rosenbergii*. *Dev Comp Immunol*. 2020;104:103564.
473. Shekhar MS, Karthic K, Kumar KV, et al. Comparative analysis of shrimp (*Penaeus vannamei*) miRNAs expression profiles during WSSV

- infection under experimental conditions and in pond culture. *Fish Shellfish Immunol.* 2019;93:288-295.
474. Yu Y, Liu JW, Li FH, Zhang XJ, Zhang CS, Xiang JH. Gene set based association analyses for the WSSV resistance of Pacific white shrimp *Litopenaeus vannamei*. *Sci Rep.* 2017;7:40549.
475. Hasanuzzaman AFM, Robledo D, Gomez-Tato A, et al. Transcriptomic profile of Manila clam (*Ruditapes philippinarum*) haemocytes in response to *Perkinsus olseni* infection. *Aquaculture.* 2017;467:170-181.
476. Hasanuzzaman AM, Robledo D, Gomez-Tato A, et al. De novo transcriptome assembly of *Perkinsus olseni* trophozoite stimulated in vitro with Manila clam (*Ruditapes philippinarum*) plasma. *J Invertebr Pathol.* 2016;135:22-33.
477. Ren YP, Xue JL, Yang HH, Pan BP, Bu WJ. Transcriptome analysis of *Ruditapes philippinarum* hepatopancreas provides insights into immune signaling pathways under vibrio anguillarum infection. *Fish Shellfish Immunol.* 2017;64:14-23.
478. Rodrigues S, Paillard C, Van Dillen S, et al. Relation between biofilm and virulence in *Vibrio tapetis*: a transcriptomic study. *Pathogens.* 2018;7(4):92.
479. Pierron F, Gonzalez P, Bertucci A, et al. Transcriptome-wide analysis of wild Asari (= Manila) clams affected by the Brown muscle disease: etiology and impacts of the disease. *Fish Shellfish Immunol.* 2019;86:179-185.
480. Yu MJ, Zheng L, Wang XB, et al. Comparative transcriptomic analysis of surf clams (*Paphia undulate*) infected with two strains of vibrio spp. reveals the identity of key immune genes involved in host defense. *BMC Genomics.* 2019;20(1):988, 988.
481. Liu ZQ, Wang LL, Zhou Z, et al. Transcriptomic analysis of oyster *Crassostrea gigas* larvae illustrates the response patterns regulated by catecholaminergic system upon acute heat and bacterial stress. *Dev Comp Immunol.* 2017;73:52-60.
482. Wang MQ, Liu M, Wang BJ, et al. Transcriptomic analysis of exosomal shuttle mRNA in Pacific oyster *Crassostrea gigas* during bacterial stimulation. *Fish Shellfish Immunol.* 2018;74:540-550.
483. Ma LP, Su LJ, Liu H, Zhao F, Zhou DQ, Duan DL. Norovirus contamination and the glycosphingolipid biosynthesis pathway in Pacific oyster: a transcriptomics study. *Fish Shellfish Immunol.* 2017;66:26-34.
484. Rosani U, Venier P. Oyster RNA-seq data support the development of Malacoherpesviridae genomics. *Front Microbiol.* 2017;8:1515.
485. Delisle L, Pualetto M, Vidal-Dupiol J, et al. High temperature induces transcriptomic changes in *Crassostrea gigas* that hinder progress of ostreid herpesvirus (OsHV-1) and promote survival. *J Exp Biol.* 2020;223(20):jeb226233.
486. Wang ZL, Wang B, Chen G, et al. Transcriptome analysis of the pearl oyster (*Pinctada fucata*) hemocytes in response to *Vibrio alginolyticus* infection. *Gene.* 2016;575(2):421-428.
487. Neave MJ, Corbeil S, McColl KA, Crane MS. Investigating the natural resistance of Blackfoot Paua *Haliotis iris* to abalone viral ganglionneuritis using whole transcriptome analysis. *Dis Aquat Organ.* 2019;135(2):107-119.
488. Bai CM, Zhang SM, Li YN, Xin LS, Rosani U, Wang CM. Dual transcriptomic analysis reveals a delayed antiviral response of *Haliotis diversicolor* supertexta against Haliotid Herpesvirus-1. *Viruses.* 2019;11(4):383.
489. Bai CM, Rosani U, Li YN, Zhang SM, Xin LS, Wang CM. RNA-seq of HaHV-1-infected abalones reveals a common transcriptional signature of Malacoherpesviruses. *Sci Rep.* 2019;9:938.
490. Zhao XL, Wan JJ, Fu JP, Shao YN, Lv ZM, Li CH. Identification of SNPs associated with disease resistance in juveniles of *Sinonovacula constricta* using RNA-seq and high-resolution melting analysis. *Aquaculture.* 2021;544:737109.
491. Zhao XL, Duan XM, Wang ZH, et al. Comparative transcriptome analysis of *Sinonovacula constricta* in gills and hepatopancreas in response to *Vibrio parahaemolyticus* infection. *Fish Shellfish Immunol.* 2017;67:523-535.
492. Jiang FJ, Yue X, Wang HX, Liu BZ. Transcriptome profiles of the clam *Meretrix petechialis* hepatopancreas in response to vibrio infection. *Fish Shellfish Immunol.* 2017;62:175-183.
493. Rosani U, Young T, Bai CM, Alfaro AC, Venier P. Dual analysis of virus-host interactions: the case of Ostreid herpesvirus 1 and the cupped oyster *Crassostrea gigas*. *Evolutionary Bioinformatics.* 2019;15:1176934319831305.
494. Sullivan ME, Proestou DA. Survival and transcriptomic responses to different *Perkinsus marinus* exposure methods in an eastern oyster family. *Aquaculture.* 2021;542:736831.
495. Johnson KM, Sirovy KA, Casas SM, La Peyre JF, Kelly MW. Characterizing the epigenetic and transcriptomic responses to *Perkinsus marinus* infection in the eastern oyster *Crassostrea virginica*. *Front Mar Sci.* 2020;7:598.
496. Proestou DA, Sullivan ME. Variation in global transcriptomic response to *Perkinsus marinus* infection among eastern oyster families highlights potential mechanisms of disease resistance. *Fish Shellfish Immunol.* 2020;96:141-151.
497. Espinosa EP, Corre E, Allam B. Pallial mucus of the oyster *Crassostrea virginica* regulates the expression of putative virulence genes of its pathogen *Perkinsus marinus*. *Int J Parasit.* 2014;44(5):305-317.
498. McDowell IC, Nikapitiya C, Aguiar D, Lane CE, Istrail S, Gomez-Chiarri M. Transcriptome of American oysters, *Crassostrea virginica*, in response to bacterial challenge: insights into potential mechanisms of disease resistance. *PLoS One.* 2014;9(8):e105097.
499. Lu M, Zhang PJ, Li CH, Lv ZM, Zhang WW, Jin CH. miRNA-133 augments coelomocyte phagocytosis in bacteria-challenged *Apostichopus japonicus* via targeting the TLR component of Irak-1 in vitro and in vivo. *Sci Rep.* 2015;5:12608.
500. Zhang WJ, Lv ZM, Li CH, et al. Transcriptome profiling reveals key roles of phagosome and NOD-like receptor pathway in spotting diseased Strongylocentrotus intermedius. *Fish Shellfish Immunol.* 2019;84:521-531.
501. Zhang WJ, Wang Z, Leng XF, et al. Transcriptome sequencing reveals phagocytosis as the main immune response in the pathogen-challenged sea urchin *Strongylocentrotus intermedius*. *Fish Shellfish Immunol.* 2019;94:780-791.
502. Linnarsson S, Teichmann SA. Single-cell genomics: coming of age. *Genome Biol.* 2016;17:97.
503. Denisenko E, Guo BB, Jones M, et al. Systematic assessment of tissue dissociation and storage biases in single-cell and single-nucleus RNA-seq workflows. *Genome Biol.* 2020;21(1):130.
504. Bakken TE, Hodge RD, Miller JA, et al. Single-nucleus and single-cell transcriptomes compared in matched cortical cell types. *PLoS One.* 2018;13(12):e0209648.
505. Drokhllyansky E, Smillie CS, Van Wittenbergh N, et al. The human and mouse enteric nervous system at single-cell resolution. *Cell.* 2020;182(6):1606.
506. Eraslan G, Drokhllyansky E, Anand S, et al. Single-nucleus cross-tissue molecular reference maps to decipher disease gene function. *Science.* 2022;376(6594):eabl4290.
507. Koiwai K, Koyama T, Tsuda S, et al. Single-cell RNA-seq analysis reveals penaeid shrimp hemocyte subpopulations and cell differentiation process. *Elife.* 2021;10:e66954.
508. West AC, Mizoro Y, Wood SH, et al. Immunologic profiling of the Atlantic Salmon gill by single nuclei transcriptomics. *Front Immunol.* 2021;12:669889.
509. Wu L, Gao A, Li L, Chen J, Li J, Ye J. A single-cell transcriptome profiling of anterior kidney leukocytes from Nile tilapia (*Oreochromis niloticus*). *Front Immunol.* 2021;12:783196.
510. Zhu W, Yang C, Chen X, et al. Single-cell ribonucleic acid sequencing clarifies cold tolerance mechanisms in the Pacific White shrimp (*Litopenaeus Vannamei*). *Front Genet.* 2021;12:792172.

511. Ståhl PL, Salmén F, Vickovic S, et al. Visualization and analysis of gene expression in tissue sections by spatial transcriptomics. *Science*. 2016;353(6294):78-82.
512. Stickels RR, Murray E, Kumar P, et al. Highly sensitive spatial transcriptomics at near-cellular resolution with slide-seqV2. *Nat Biotechnol*. 2021;39(3):313.
513. Vickovic S, Eraslan G, Salmen F, et al. High-definition spatial transcriptomics for in situ tissue profiling. *Nat Methods*. 2019;16(10):987-990.
514. Rodriques SG, Stickels RR, Goeva A, et al. Slide-seq: a scalable technology for measuring genome-wide expression at high spatial resolution. *Science*. 2019;363(6434):1463.
515. Longo SK, Guo MG, Ji AL, Khavari PA. Integrating single-cell and spatial transcriptomics to elucidate intercellular tissue dynamics. *Nat Rev Genet*. 2021;22(10):627-644.
516. Braden LM, Koop BF, Jones SR. Signatures of resistance to *Lepeophtheirus salmonis* include a TH2-type response at the louse-salmon interface. *Dev Comp Immunol*. 2015;48(1):178-191.
517. Paul I, White C, Turcinovic I, Emili A. Imaging the future: the emerging era of single-cell spatial proteomics. *Febs J*. 2021;288(24):6990-7001.
518. Moreira M, Schrama D, Farinha AP, et al. Fish pathology research and diagnosis in aquaculture of farmed fish; a proteomics perspective. *Animals*. 2021;11(1):125.
519. Li JM, Cai ZY, Bomgardner RD, et al. TMTpro-18plex: the expanded and complete set of TMTpro reagents for sample multiplexing. *J Proteome Res*. 2021;20(5):2964-2972.
520. Meier F, Brunner AD, Frank M, et al. diaPASEF: parallel accumulation-serial fragmentation combined with data-independent acquisition. *Nat Methods*. 2020;17(12):1229.
521. Peterson AC, Russell JD, Bailey DJ, Westphal MS, Coon JJ. Parallel reaction monitoring for high resolution and high mass accuracy quantitative, targeted proteomics. *Mol Cell Proteomics*. 2012;11(11):1475-1488.
522. Michard C, Doublet P. Post-translational modifications are key players of the *Legionella pneumophila* infection strategy. *Front Microbiol*. 2015;6:687.
523. Hu J, Zhang L, Liu XF. Role of post-translational modifications in influenza a virus life cycle and host innate immune response. *Front Microbiol*. 2020;11:517461.
524. Lin JS, Lai EM. Protein-protein interactions: co-immunoprecipitation. *Methods Mol Biol*. 2017;1615:211-219.
525. Kim DI, Jensen SC, Noble KA, et al. An improved smaller biotin ligase for BioID proximity labeling. *Mol Biol Cell*. 2016;27(8):1188-1196.
526. Liu XA, Salokas K, Weldatsadik RG, Gawryski L, Varjosalo M. Combined proximity labeling and affinity purification-mass spectrometry workflow for mapping and visualizing protein interaction networks. *Nat Protoc*. 2020;15(10):3182-3211.
527. Xiong ZR, Lo HP, McMahon KA, et al. In vivo proteomic mapping through GFP-directed proximity-dependent biotin labelling in zebrafish. *Elife*. 2021;10:e64631.
528. Heusel M, Bludau I, Rosenberger G, et al. Complex-centric proteome profiling by SEC-SWATH-MS. *Mol Syst Biol*. 2019;15(1):e8438.
529. Gorka M, Swart C, Siemiatkowska B, et al. Protein complex identification and quantitative complexome by CN-PAGE. *Sci Rep*. 2019;9:11523.
530. Liu F, Rijkers DTS, Post H, Heck AJR. Proteome-wide profiling of protein assemblies by cross-linking mass spectrometry. *Nat Methods*. 2015;12(12):1179-1184.
531. Wang C, Wei M, Wu G, et al. Proteomics analysis reveals a critical role for the WSSV immediate-early protein IE1 in modulating the host prophenoloxidase system. *Virulence*. 2022;13(1):936-948.
532. Alfaro AC, Nguyen TV, Bayot B, Leon JAR, Dominguez-Borbor C, Sonnenholzner S. Metabolic responses of whiteleg shrimp to white spot syndrome virus (WSSV). *J Invertebr Pathol*. 2021;180:107545.
533. Nguyen TV, Alfaro A, Arroyo BB, Leon JAR, Sonnenholzner S. Metabolic responses of penaeid shrimp to acute hepatopancreatic necrosis disease caused by *Vibrio parahaemolyticus*. *Aquaculture*. 2021;533:736174.
534. Mahajan S, Choudhary S, Kumar P, Tomar S. Antiviral strategies targeting host factors and mechanisms obliging plus ssRNA viral pathogens. *Bioorg Med Chem*. 2021;46:116356.
535. Selander E, Heuschele J, Larsson AI. Hydrodynamic properties and distribution of bait downstream of a zooplankton trap. *J Plankton Res*. 2017;39(6):1020-1027.
536. Kuhlisch C, Pohnert G. Metabolomics in chemical ecology. *Nat Prod Rep*. 2015;32(7):937-955.
537. Fields DM, Skiftesvik AB, Browman HI. Behavioural responses of infective-stage copepodids of the salmon louse (*Lepeophtheirus salmonis*, Copepoda:Caligidae) to host-related sensory cues. *J Fish Dis*. 2018;41(6):875-884.
538. Flamarique IN, Browman HI, Belanger M, Boxaspen K. Ontogenetic changes in visual sensitivity of the parasitic salmon louse *Lepeophtheirus salmonis*. *J Exp Biol*. 2000;203(11):1649-1657.
539. Gotoh O. Evolution of cytochrome p450 genes from the viewpoint of genome informatics. *Biol Pharm Bull*. 2012;35(6):812-817.
540. Lewis DF, Watson E, Lake BG. Evolution of the cytochrome P450 superfamily: sequence alignments and pharmacogenetics. *Mutat Res*. 1998;410(3):245-270.
541. Yang YH, Wang JL, Miranda CL, Buhler DR. CYP2M1: cloning, sequencing, and expression of a new cytochrome P450 from rainbow trout liver with fatty acid (omega-6)-hydroxylation activity. *Arch Biochem Biophys*. 1998;352(2):271-280.
542. Krasnov A, Timmerhaus G, Afanasyev S, Jørgensen SM. Development and assessment of oligonucleotide microarrays for Atlantic salmon (*Salmo salar* L.). Comparative biochemistry and physiology part D. *Genomics & Proteomics*. 2011;6(1):31-38.
543. Cong L, Ran FA, Cox D, et al. Multiplex genome engineering using CRISPR/Cas systems. *Science*. 2013;339(6121):819-823.
544. Gilbert LA, Larson MH, Morsut L, et al. CRISPR-mediated modular RNA-guided regulation of transcription in eukaryotes. *Cell*. 2013;154(2):442-451.
545. Bae T, Hur JW, Kim D, Hur JK. Recent trends in CRISPR-Cas system: genome, epigenome, and transcriptome editing and CRISPR delivery systems. *Genes Genomics*. 2019;41(8):871-877.
546. Uusi-Makela MIE, Barker HR, Bauerlein CA, Hakkinen T, Nykter M, Ramet M. Chromatin accessibility is associated with CRISPR-Cas9 efficiency in the zebrafish (*Danio rerio*). *PLoS One*. 2018;13(4):e0196238.
547. Dunham RA, Warr GW, Nichols A, et al. Enhanced bacterial disease resistance of transgenic channel catfish *Ictalurus punctatus* possessing cecropin genes. *Marine Biotechnol*. 2002;4(3):338-344.
548. Elasad A, Khalil K, Cline D, et al. Microinjection of CRISPR/Cas9 protein into channel catfish, *Ictalurus punctatus*, embryos for gene editing. *J Vis Exp*. 2018;131:56275.
549. Simora RMC, Xing D, Bangs XR, et al. CRISPR/Cas9-mediated knock-in of alligator cathelicidin gene in a non-coding region of channel catfish genome. *Sci Rep*. 2020;10(1):22271.
550. Ma J, Fan YD, Zhou Y, et al. Efficient resistance to grass carp reovirus infection in JAM-A knockout cells using CRISPR/Cas9. *Fish Shellfish Immunol*. 2018;76:206-215.
551. Zhong JY, Wang YP, Zhu ZY. Introduction of the human lactoferrin gene into grass carp (*Ctenopharyngodon idellus*) to increase resistance against GCH virus. *Aquaculture*. 2002;214(1-4):93-101.
552. Mao WF, Wang YP, Wang WB, Bo W, Feng JX, Zhu ZY. Enhanced resistance to *Aeromonas hydrophila* infection and enhanced phagocytic activities in human lactoferrin-transgenic grass carp (*Ctenopharyngodon idellus*). *Aquaculture*. 2004;242(1-4):93-103.
553. Chakrapani V, Patra SK, Panda RP, Rasal KD, Jayasankar P, Barman HK. Establishing targeted carp TLR22 gene disruption via

- homologous recombination using CRISPR/Cas9. *Dev Comp Immunol*. 2016;61:242-247.
554. Chiou PP, Chen MJ, Lin CM, et al. Production of homozygous transgenic rainbow trout with enhanced disease resistance. *Marine Biotechnol*. 2014;16(3):299-308.
555. Sun YY, Zhang JQ, Xiang JH. A CRISPR/Cas9-mediated mutation in chitinase changes immune response to bacteria in *Exopalaemon carinicauda*. *Fish Shellfish Immunol*. 2017;71:43-49.
556. Datsomor AK, Olsen RE, Zic N, et al. CRISPR/Cas9-mediated editing of Delta 5 and Delta 6 desaturases impairs Delta 8-desaturation and docosahexaenoic acid synthesis in Atlantic salmon (*Salmo salar* L.). *Sci Rep*. 2019;9:16888.
557. Datsomor AK, Zic N, Li KS, et al. CRISPR/Cas9-mediated ablation of elovl2 in Atlantic salmon (*Salmo salar* L.) inhibits elongation of polyunsaturated fatty acids and induces Srebp-1 and target genes. *Sci Rep*. 2019;9:7533.
558. Straume AH, Kjaerner-Semb E, Skaftnesmo KO, et al. Single nucleotide replacement in the Atlantic salmon genome using CRISPR/Cas9 and asymmetrical oligonucleotide donors. *BMC Genomics*. 2021;22(1):563.
559. Straume AH, Kjaerner-Semb E, Skaftnesmo KO, et al. Indel locations are determined by template polarity in highly efficient in vivo CRISPR/Cas9-mediated HDR in Atlantic salmon. *Sci Rep*. 2020;10(1):409.
560. Edvardsen RB, Leininger S, Kleppe L, Skaftnesmo KO, Wargelius A. Targeted mutagenesis in Atlantic Salmon (*Salmo salar* L.) using the CRISPR/Cas9 system induces complete knockout individuals in the F0 generation. *PLoS One*. 2014;9(9):e108622.
561. Gratacap RL, Jin YH, Mantopoulou M, Houston RD. Efficient genome editing in multiple salmonid cell lines using ribonucleoprotein complexes. *Marine Biotechnol*. 2020;22(5):717-724.
562. Gratacap RL, Regan T, Dehler CE, et al. Efficient CRISPR/Cas9 genome editing in a salmonid fish cell line using a lentivirus delivery system. *BMC Biotechnol*. 2020;20(1):35.
563. Gonzalez-Duarte RJ, de Aquino WE, Garcia-Carreno F, Rojo-Arreola L. The hurdles of delivery CRISPR-Cas9 components for genome editing in penaeid shrimps. *Aquacult Res*. 2021;52:5297-5306.
564. Gui TS, Zhang JQ, Song FG, et al. CRISPR/Cas9-mediated genome editing and mutagenesis of EcChi4 in *Exopalaemon carinicauda*. *G3-Genes Genomes Genet*. 2016;6(11):3757-3764.
565. Sun YY, Liu MF, Yan CC, et al. CRISPR/Cas9-mediated deletion of beta, beta-carotene 9', 10'-oxygenase gene (EcBCO2) from *Exopalaemon carinicauda*. *Int J Biol Macromol*. 2020;151:168-177.
566. Sun YY, Yan CC, Liu MF, et al. CRISPR/Cas9-mediated deletion of one carotenoid isomeroxygenase gene (EcNinaB-X1) from *Exopalaemon carinicauda*. *Fish Shellfish Immunol*. 2020;97:421-431.
567. Zhang JQ, Song FG, Sun YY, Yu KJ, Xiang JH. CRISPR/Cas9-mediated deletion of EcMIH shortens metamorphosis time from mysis larva to postlarva of *Exopalaemon carinicauda*. *Fish Shellfish Immunol*. 2018;77:244-251.
568. Gao Y, Zhang XX, Zhang XJ, Yuan JB, Xiang JH, Li FH. CRISPR/Cas9-mediated mutation reveals Pax6 is essential for development of the compound eye in Decapoda *Exopalaemon carinicauda*. *Dev Biol*. 2020;465(2):157-167.
569. Burkard C, Lillico SG, Reid E, et al. Precision engineering for PRRSV resistance in pigs: macrophages from genome edited pigs lacking CD163 SRCR5 domain are fully resistant to both PRRSV genotypes while maintaining biological function. *PLoS Pathog*. 2017;13(2):28.
570. Ren XC, Liu QH. LvCPG2 facilitated WSSV infection by interaction with VP26 and VP28. *Fish Shellfish Immunol*. 2021;118:313-320.
571. Kao ZN, Liu CH, Liu WJ, Kumar R, Leu JH, Wang HC. Shrimp SIRT1 activates of the WSSV IE1 promoter independently of the NF-kappa B binding site. *Fish Shellfish Immunol*. 2020;106:910-919.
572. Sanson KR, Hanna RE, Hegde M, et al. Optimized libraries for CRISPR-Cas9 genetic screens with multiple modalities. *Nat Commun*. 2018;9(1):5416.
573. Goddard ME, Hayes BJ. Genomic selection. *J Anim Breed Genet*. 2007;124:323-330.
574. Vallejo RL, Cheng H, Fragomeni BO, et al. The accuracy of genomic predictions for bacterial cold water disease resistance remains higher than the pedigree-based model one generation after model training in a commercial rainbow trout breeding population. *Aquaculture*. 2021;545:737164.
575. Vallejo RL, Silva RMO, Evenhuis JP, et al. Accurate genomic predictions for BCWD resistance in rainbow trout are achieved using low-density SNP panels: evidence that long-range LD is a major contributing factor. *J Anim Breed Genet*. 2018;135(4):263-274.
576. Tsai H-Y, Matika O, Edwards SM, et al. Genotype imputation to improve the cost-efficiency of genomic selection in farmed Atlantic Salmon. *G3 Genes Genomes Genetics*. 2017;7(4):1377-1383.
577. Yoshida GM, Bangera R, Carvalheiro R, et al. Genomic prediction accuracy for resistance against *Piscirickettsia salmonis* in farmed rainbow trout. *G3-Genes Genomes Genet*. 2018;8(2):719-726.
578. Yoshida GM, Carvalheiro R, Rodriguez FH, Lhorente JP, Yanez JM. Single-step genomic evaluation improves accuracy of breeding value predictions for resistance to infectious pancreatic necrosis virus in rainbow trout. *Genomics*. 2019;111(2):127-132.
579. Vallejo RL, Fragomeni BO, Cheng H, et al. Assessing accuracy of genomic predictions for resistance to infectious hematopoietic necrosis virus with progeny testing of selection candidates in a commercial rainbow trout breeding population. *Front Vet Sci*. 2020;7:590048.
580. Griot R, Allal F, Phocas F, et al. Optimization of genomic selection to improve disease resistance in two marine fishes, the European Sea bass (*Dicentrarchus labrax*) and the Gilthead Sea bream (*Sparus aurata*). *Front Genet*. 2021;12:665920.
581. Paliokostas C, Ferrareso S, Franch R, Houston RD, Bargelloni L. Genomic prediction of resistance to Pasteurellosis in Gilthead Sea bream (*Sparus aurata*) using 2b-RAD sequencing. *G3-Genes Genomes Genet*. 2016;6(11):3693-3700.
582. Oikonomou S, Samaras A, Tekeoglou M, et al. Genomic selection and genome-wide association analysis for stress response, disease resistance and body weight in European seabass. *Animals (Basel)*. 2022;12(3):277.
583. Vu NT, Phuc TH, Oanh KTP, Sang NV, Trang TT, Nguyen NH. Accuracies of genomic predictions for disease resistance of striped catfish to *Edwardsiella ictaluri* using artificial intelligence algorithms. *G3 (Bethesda)*. 2022;12(1):jkab361.
584. Lin Z, Hosoya S, Sato M, et al. Genomic selection for heterobothriosis resistance concurrent with body size in the tiger pufferfish, *Takifugu rubripes*. *Sci Rep*. 2020;10(1):19976.
585. Lillehammer M, Bangera R, Salazar M, et al. Genomic selection for white spot syndrome virus resistance in whiteleg shrimp boosts survival under an experimental challenge test. *Sci Rep*. 2020;10(1):20571.
586. Gutierrez AP, Symonds J, King N, Steiner K, Bean TP, Houston RD. Potential of genomic selection for improvement of resistance to ostreid herpesvirus in Pacific oyster (*Crassostrea gigas*). *Anim Genet*. 2020;51(2):249-257.
587. Lu S, Liu Y, Yu XJ, et al. Prediction of genomic breeding values based on pre-selected SNPs using ssGBLUP, WssGBLUP and BayesB for Edwardsiellosis resistance in Japanese flounder. *Genet Sel Evol*. 2020;52(1):49.
588. Lillehammer M, Meuwissen TH, Sonesson A. A low-marker density implementation of genomic selection in aquaculture using within-family genomic breeding values. *Genet Sel Evol*. 2013;45(1):39.
589. Nielsen H, Sonesson A, Yazdi H, Meuwissen T. Comparison of accuracy of genome-wide and BLUP breeding value estimates in sib based aquaculture breeding schemes. *Aquaculture*. 2009;289:259-264.

590. Sonesson A, Meuwissen T, Goddard M. The use of communal rearing of families and DNA pooling in aquaculture genomic selection schemes. *Genet Sel Evol.* 2010;42:41.
591. Tsairidou S, Hamilton A, Robledo D, Bron JE, Houston RD. Optimizing low-cost genotyping and imputation strategies for genomic selection in Atlantic Salmon. *G3 Genes|Genomes|Genetics.* 2020;10(2):581-590.
592. Kriaridou C, Tsairidou S, Houston RD, Robledo D. Genomic prediction using low density marker panels in aquaculture: performance across species, traits, and genotyping platforms. *Front Genet.* 2020; 11:124.
593. VanRaden P. Efficient methods to compute genomic predictions. *J Dairy Sci.* 2008;91:4414-4423.
594. Legarra A, Aguilar I, Misztal I. A relationship matrix including full pedigree and genomic information. *J Dairy Sci.* 2009;92:4656-4663.
595. Raoul J, Swan AA, Elsen JM. Using a very low-density SNP panel for genomic selection in a breeding program for sheep. *Genet Sel Evol.* 2017;49:76.
596. Robledo D, Palaiokostas C, Bargelloni L, Martinez P, Houston R. Applications of genotyping by sequencing in aquaculture breeding and genetics. *Rev Aquac.* 2018;10(3):670-682.
597. Houston RD, Bean TP, Macqueen DJ, et al. Harnessing genomics to fast-track genetic improvement in aquaculture. *Nat Rev Genet.* 2020;21(7):389-409.
598. Gratacap RL, Wargelius A, Edvardson RB, Houston RD. Potential of genome editing to improve aquaculture breeding and production. *Trends Genet.* 2019;35(9):672-684.
599. Jin YH, Robledo D, Hickey JM, McGrew MJ, Houston RD. Surrogate broodstock to enhance biotechnology research and applications in aquaculture. *Biotechnol Adv.* 2021;49:107756.
600. Fernandez-Montero A, Torrecillas S, Acosta F, et al. Improving greater amberjack (*Seriola dumerilli*) defenses against monogenean parasite *Neobenedenia girellae* infection through functional dietary additives. *Aquaculture.* 2021;534:736317.
601. Adeshina I, Abdel-Tawwab M, Tijjani ZA, Tihamiyu LO, Jahanbakhshi A. Dietary *Tridax procumbens* leaves extract stimulated growth, antioxidants, immunity, and resistance of *Nile tilapia, Oreochromis niloticus*, to monogenean parasitic infection. *Aquaculture.* 2021;532:736047.
602. Xueqin J, Kania PW, Buchmann K. Comparative effects of four feed types on white spot disease susceptibility and skin immune parameters in rainbow trout, *Oncorhynchus mykiss* (Walbaum). *J Fish Dis.* 2012;35(2):127-135.
603. Harikrishnan R, Kim JS, Kim MC, Balasundaram C, Heo MS. *Kalopanax pictum* as feed additive controls bacterial and parasitic infections in kelp grouper, *Epinephelus bruneus*. *Fish Shellfish Immunol.* 2011;31(6):801-807.
604. O'Shea B, Wadsworth S, Marambio JP, Birkett MA, Pickett JA, Mordue AJ. Disruption of host-seeking behaviour by the salmon louse, *Lepeophtheirus salmonis*, using botanically derived repellents. *J Fish Dis.* 2017;40(4):495-505.
605. Nilsen F. *Annual report 2015.* Sea Lice Research Centre; 2015.
606. Nilsen F. *Annual report 2017.* Sea Lice Research Centre; 2017.
607. Hastie LC, Wallace C, Birkett MA, et al. Prevalence and infection intensity of sea lice (*Lepeophtheirus salmonis*) on Atlantic salmon (*Salmo salar*) host is reduced by the non-host compound 2-aminoacetophenone. *Aquaculture.* 2013;410:179-183.
608. Aida V, Pliasis VC, Neasham PJ, et al. Novel vaccine technologies in veterinary medicine: a herald to human medicine vaccines. *Front Vet Science.* 2021;8:654289.
609. Karlsen M, Villoing S, Rimstad E, Nylund A. Characterization of untranslated regions of the salmonid alphavirus 3 (SAV3) genome and construction of a SAV3 based replicon. *Virology.* 2009;6:173.
610. Wolf A, Hodneland K, Frost P, Braaen S, Rimstad E. A hemagglutinin-esterase-expressing salmonid alphavirus replicon protects Atlantic salmon (*Salmo salar*) against infectious salmon anemia (ISA). *Vaccine.* 2013;31(4):661-669.
611. Wolf A, Hodneland K, Frost P, Hoeijmakers M, Rimstad E. Salmonid alphavirus-based replicon vaccine against infectious salmon anemia (ISA): impact of immunization route and interactions of the replicon vector. *Fish Shellfish Immunol.* 2014;36(2):383-392.
612. Grayson TH, John RJ, Wadsworth S, et al. Immunization of Atlantic salmon against the salmon louse: identification of antigens and effects on louse fecundity. *J Fish Biol.* 1995;47:85-94.
613. Tartor H, Karlsen M, Skern-Mauritzen R, et al. Protective immunization of Atlantic Salmon (*Salmo salar* L.) against Salmon lice (*Lepeophtheirus salmonis*) infestation. *Vaccine.* 2022;10(1):16.
614. Contreras M, Karlsen M, Villar M, et al. Vaccination with Ectoparasite proteins involved in midgut function and blood digestion reduces Salmon louse infestations. *Vaccine.* 2020;8(1):32.
615. Swain JK, Carpio Y, Johansen LH, et al. Impact of a candidate vaccine on the dynamics of salmon lice (*Lepeophtheirus salmonis*) infestation and immune response in Atlantic salmon (*Salmo salar* L.). *PLoS One.* 2020;15(10):e0239827.
616. Leal Y, Velazquez J, Hernandez L, et al. Promiscuous T cell epitopes boosts specific IgM immune response against a PO peptide antigen from sea lice in different teleost species. *Fish Shellfish Immunol.* 2019;92:322-330.
617. Carpio Y, Basabe L, Acosta J, et al. Novel gene isolated from *Caligus rogercresseyi*: a promising target for vaccine development against sea lice. *Vaccine.* 2011;29(15):2810-2820.
618. Carpio Y, Garcia C, Pons T, et al. Akinins in sea lice: first steps towards a deeper understanding. *Exp Parasitol.* 2013;135(2): 188-199.
619. Valenzuela-Munoz V, Benavente BP, Casuso A, et al. Transcriptome and morphological analysis in *Caligus rogercresseyi* uncover the effects of Atlantic salmon vaccination with IPath (R). *Fish Shellfish Immunol.* 2021;117:169-178.
620. Contreras M, Villar M, de la Fuente J. A Vaccinomics approach for the identification of tick protective antigens for the control of *Ixodes ricinus* and *Dermacentor reticulatus* infestations in companion animals. *Front Physiol.* 2019;10:977.
621. de la Fuente J, Rodriguez M, Montero C, et al. Vaccination against ticks (*Boophilus* spp.): the experience with the Bm86-based vaccine Gavac (TM). *Genet Anal.* 1999;15(3-5):143-148.
622. Ye T, Wu XT, Wu WL, Dai CJ, Yuan JJ. Ferritin protect shrimp *Litopenaeus vannamei* from WSSV infection by inhibiting virus replication. *Fish Shellfish Immunol.* 2015;42(1):138-143.
623. Dekham K, Jitrakorn S, Charoonnart P, Isarangkul D, Chaturongakul S, Saksmerprom V. Probiotics expressing double-stranded RNA targeting VP28 efficiently protect shrimps from WSSV infection. *Aquac Rep.* 2022;23:101067.
624. Marr EJ, Sargison ND, Nisbet AJ, Burgess STG. RNA interference for the identification of ectoparasite vaccine candidates. *Parasite Immunol.* 2014;36(11):614-624.
625. Dalvin S, Frost P, Biering E, et al. Functional characterisation of the maternal yolk-associated protein (LsYAP) utilising systemic RNA interference in the salmon louse (*Lepeophtheirus salmonis*) (Crustacea: Copepoda). *Int J Parasit.* 2009;39(13):1407-1415.
626. Trosse C, Nilsen F, Dalvin S. RNA interference mediated knockdown of the KDEL receptor and COPB2 inhibits digestion and reproduction in the parasitic copepod *Lepeophtheirus salmonis*. *Comp Biochem Physiol B-Biochem Molec Biol.* 2014;170:1-9.
627. Komisarczuk AZ, Kongshaug H, Li M, Nilsen F. RNAi mediated myosuppressin deficiency affects muscle development and survival in the salmon louse (*Lepeophtheirus salmonis*). *Sci Rep.* 2019;9:6944.
628. Campbell EM, Pert CC, Bowman AS. RNA-interference methods for gene-knockdown in the sea louse, *Lepeophtheirus salmonis*: studies on a putative prostaglandin E synthase. *Parasitology.* 2009;136(8): 867-874.

629. Kvamme BO, Skern R, Frost P, Nilsen F. Molecular characterisation of five trypsin-like peptidase transcripts from the salmon louse (*Lepeophtheirus salmonis*) intestine. *Int J Parasit.* 2004;34(7):823-832.
630. Iversen A, Hermansen Ø, Nystøyl R, Hess EJ. Kostnadsutvikling i lakseoppdrett—med fokus på før- og lusekostnader. Nofima Rapport 24–2017. 2017.
631. Champer J, Yang E, Lee E, Liu J, Clark AG, Messer PW. A CRISPR homing gene drive targeting a haplolethal gene removes resistance alleles and successfully spreads through a cage population. *Proc Natl Acad Sci.* 2020;117(39):24377-24383.
632. Hulst AD, de Jong MCM, Bijma P. Why genetic selection to reduce the prevalence of infectious diseases is way more promising than currently believed. *Genetics.* 2021;217(4):iyab024.
633. Doeschl-Wilson A, Knap PW, Opiressnig T, More SJ. Review: livestock disease resilience: from individual to herd level. *Animal.* 2021; 15:100286.
634. Prentice JC, Pooley CM, Tsairidou S, et al. Transmission experiment in turbot shows highgenetic variation in host infectivity affecting disease spread and survival. *Proceedings of the 12th World Congress on Genetics Applied to Livestock Production, 2022, Rotterdam: WCGALP.*
635. Tsairidou S, Anacleto O, Woolliams JA, Doeschl-Wilson A. Enhancing genetic disease control by selecting for lower host infectivity and susceptibility. *Heredity.* 2019;122(6):742-758.
636. Stormoen M, Skjerve E, Aunsmo A. Modelling salmon lice, *Lepeophtheirus salmonis*, reproduction on farmed Atlantic salmon, *Salmo salar* L. *J Fish Dis.* 2013;36(1):25-33.
637. Brattsten LB, Holyoke CW, Leeper JR, Raffa KF. Insecticide resistance—challenge to pest-management and basic research. *Science.* 1986;231(4743):1255-1260.
638. Holt JS, Lebaron HM. Significance and distribution of herbicide resistance. *Weed Technol.* 1990;4(1):141-149.
639. Kaplan RM, Vidyashankar AN. An inconvenient truth: global worming and anthelmintic resistance. *Vet Parasitol.* 2012;186(1-2):70-78.
640. Tabashnik BE, Carriere Y. Surge in insect resistance to transgenic crops and prospects for sustainability. *Nat Biotechnol.* 2017;35(10): 926-935.
641. Fjørtoft HB, Nilsen F, Besnier F, et al. Aquaculture-driven evolution: distribution of pyrethroid resistance in the salmon louse throughout the North Atlantic in the years 2000–2017. *ICES J Marine Sci.* 2020; 77(5):1806-1815.
642. Besnier F, Kent M, Skern-Mauritzen R, et al. Human-induced evolution caught in action: SNP-array reveals rapid amphi-Atlantic spread of pesticide resistance in the salmon ectoparasite *Lepeophtheirus salmonis*. *BMC Genomics.* 2014;15:937.
643. Kaur K, Besnier F, Glover KA, et al. The mechanism (Phe362Tyr mutation) behind resistance in *Lepeophtheirus salmonis* pre-dates organophosphate use in salmon farming. *Sci Rep.* 2017;7:12349.
644. Allen RL, Bittner-Eddy PD, Grenvitt-Briggs LJ, et al. Host-parasite coevolutionary conflict between *Arabidopsis* and downy mildew. *Science.* 2004;306(5703):1957-1960.
645. Cowger C, Hoffer ME, Mundt CC. Specific adaptation by *Mycosphaerella graminicola* to a resistant wheat cultivar. *Plant Pathol.* 2000;49(4):445-451.
646. Park RF. Breeding cereals for rust resistance in Australia. *Plant Pathol.* 2008;57(4):591-602.
647. Rouxel T, Penaud A, Pinochet X, et al. A 10-year survey of populations of *Leptosphaeria maculans* in France indicates a rapid adaptation towards the Rlm1 resistance gene of oilseed rape. *Eur J Plant Pathol.* 2003;109(8):871-881.
648. Gjerde B. Er avl mot lus eit sisyfosarbeid? *Norsk Fiskoppdrett.* 2018; 8:90.
649. Stear MJ, Bishop SC, Mallard BA, Raadsma H. The sustainability, feasibility and desirability of breeding livestock for disease resistance. *Res Vet Sci.* 2001;71(1):1-7.
650. Kerr PJ, Cattadori IM, Liu J, et al. Next step in the ongoing arms race between myxoma virus and wild rabbits in Australia is a novel disease phenotype. *Proc Natl Acad Sci U S A.* 2017;114(35):9397-9402.
651. Mone Y, Ribou AC, Cosseau C, et al. An example of molecular coevolution: reactive oxygen species (ROS) and ROS scavenger levels in *Schistosoma mansoni*/*Biomphalaria glabrata* interactions. *Int J Parasit.* 2011;41(7):721-730.
652. Webster JP, Gower CM, Blair L. Do hosts and parasites coevolve? Empirical support from the *Schistosoma* system. *Am Nat.* 2004;164 (5):S33-S51.
653. Wilfert L, Jiggins FM. Disease association mapping in *Drosophila* can be replicated in the wild. *Biol Lett.* 2010;6(5):666-668.
654. Kemper KE, Goddard ME, Bishop SC. Adaptation of gastrointestinal nematode parasites to host genotype: single locus simulation models. *Genet Sel Evol.* 2013;45:14.
655. Skern-Mauritzen R, Torrissen O, Glover KA. Pacific and Atlantic *Lepeophtheirus salmonis* (Kroyer, 1838) are allopatric subspecies: *Lepeophtheirus salmonis salmonis* and *L. salmonis oncorhynchi* subspecies novo. *BMC Genet.* 2014;15:32.
656. Yazawa R, Yasuike M, Leong J, et al. EST and mitochondrial DNA sequences support a distinct Pacific form of Salmon louse, *Lepeophtheirus salmonis*. *Marine Biotechnol.* 2008;10(6):741-749.
657. Beamish RJ, Neville CM, Sweeting RM, Ambers N. Sea lice on adult Pacific salmon in the coastal waters of Central British Columbia, Canada. *Fish Res.* 2005;76(2):198-208.
658. Gottesfeld AS, Proctor B, Rolston LD, Carr-Harris C. Sea lice, *Lepeophtheirus salmonis*, transfer between wild sympatric adult and juvenile salmon on the north coast of British Columbia, Canada. *J Fish Dis.* 2009;32(1):45-57.
659. Nagasawa K. Annual changes in the population size of the salmon louse *Lepeophtheirus salmonis* (Copepoda: Caligidae) on high-seas Pacific Salmon (*Oncorhynchus* spp.), and relationship to host abundance. *Hydrobiologia.* 2001;453(1-3):411-416.
660. Jones S, Kim E, Dawe S. Experimental infections with *Lepeophtheirus salmonis* (Kroyer) on threespine sticklebacks, *Gasterosteus aculeatus* L., and juvenile Pacific salmon, *Oncorhynchus* spp. *J Fish Dis.* 2006; 29(8):489-495.
661. Jones SRM, Proserpi-Porta G, Kim E, Callow P, Hargreaves NB. The occurrence of *Lepeophtheirus salmonis* and *Caligus clemensi* (Copepoda: Caligidae) on three-spine stickleback *Gasterosteus aculeatus* in coastal British Columbia. *J Parasitol.* 2006;92(3):473-480.
662. Bishop SC, Chesnais J, Stear MJ. *Breeding for disease resistance: issues and opportunities.* 7th World Congress on Genetic Applies to Livestock Production. Montpellier; 2002.
663. Kiyosawa S. Genetics and epidemiological modeling of breakdown of plant-disease resistance. *Annu Rev Phytopathol.* 1982;20: 93-117.
664. Rimbaud L, Papaix J, Barrett LG, Burdon JJ, Thrall PH. Mosaics, mixtures, rotations or pyramiding: what is the optimal strategy to deploy major gene resistance? *Evol Appl.* 2018;11(10):1791-1810.
665. Kreitzman M, Ashander J, Driscoll J, et al. Wild Salmon sustain the effectiveness of parasite control on Salmon farms: conservation implications from an evolutionary ecosystem service. *Conserv Lett.* 2018;11(2):e12395.
666. McEwan GF, Groner ML, Fast MD, Gettinby G, Revie CW. Using agent-based modelling to predict the role of wild Refugia in the evolution of resistance of sea lice to Chemotherapeutants. *PLoS One.* 2015;10(10):23.
667. Crowder DW, Carriere Y. Comparing the refuge strategy for managing the evolution of insect resistance under different reproductive strategies. *J Theor Biol.* 2009;261(3):423-430.
668. Bateman AW, Peacock SJ, Krkosek M, Lewis MA. Migratory hosts can maintain the high-dose/refuge effect in a structured host-parasite system: the case of sea lice and salmon. *Evol Appl.* 2020;13(10): 2521-2535.

669. Coates AL. The evolution of salmon lice (*Lepeophtheirus salmonis*) in response to parasite management strategies in salmon aquaculture. PhD Thesis. 2022, Australia: The University of Melbourne.
670. Onstad DW, Liu XX, Chen M, Roush R, Shelton AM. Modeling the integration of parasitoid, insecticide, and transgenic insecticidal crop for the long-term control of an insect pest. *J Econ Entomol*. 2013; 106(3):1103-1111.
671. Yang Z, Yu Y, Tay YX, Yue GH. Genome editing and its applications in genetic improvement in aquaculture. *Rev Aquac*. 2021;14(1):178-191.
672. Lyzenga WJ, Pozniak CJ, Kagale S. Advanced domestication: harnessing the precision of gene editing in crop breeding. *Plant Biotechnol J*. 2021;19(4):660-670.
673. Bastiaansen JWM, Bovenhuis H, Groenen MAM, Megens HJ, Mulder HA. The impact of genome editing on the introduction of monogenic traits in livestock. *Genet Sel Evol*. 2018;50(1):18.
674. Gonen S, Jenko J, Gorjanc G, Mileham AJ, Whitelaw CB, Hickey JM. Potential of gene drives with genome editing to increase genetic gain in livestock breeding programs. *Genet Sel Evol*. 2017;49(1):3.
675. Jenko J, Gorjanc G, Cleveland MA, et al. Potential of promotion of alleles by genome editing to improve quantitative traits in livestock breeding programs. *Genet Sel Evol*. 2015;47:55.
676. Petersen GEL, Buntjer JB, Hely FS, Byrne TJ, Doeschl-Wilson A. Modeling suggests gene editing combined with vaccination could eliminate a persistent disease in livestock. *Proc Natl Acad Sci*. 2022; 119(9):e2107224119.
677. Blix TB, Dalmo RA, Wargelius A, Myhr AI. Genome editing on finfish: current status and implications for sustainability. *Rev Aquac*. 2021; 13(4):2344-2363.
678. Olesen I, Myhr AI, Rosendal GK. Sustainable aquaculture: are we getting there? Ethical perspectives on Salmon farming. *J Agric Environ Ethics*. 2011;24(4):381-408.
679. Okoli AS, Blix T, Myhr AI, Xu W, Xu X. Sustainable use of CRISPR/Cas in fish aquaculture: the biosafety perspective. *Transgenic Res*. 2021;31(1):1-21.
680. Torstensen B. *RRI Responsible Use of Gene Editing in Research and Innovation in Nofima*. Nofima; 2019:4647.
681. Forsberg E-M. The ethical matrix—a tool for ethical assessments of biotechnology. *Global Bioethics*. 2004;17(1):167-172.
682. Mepham B, Kaiser M, Thorstensen E, Tomkins S, Millar K. *Ethical Matrix Manual*. LEI, onderdeel van Wageningen UR; 2006.

How to cite this article: Robinson NA, Robledo D, Sveen L, et al. Applying genetic technologies to combat infectious diseases in aquaculture. *Rev Aquac*. 2023;15(2):491-535. doi:[10.1111/raq.12733](https://doi.org/10.1111/raq.12733)