Leveraging CRISPR-Cas9 to Illuminate Zoonotic Pathways of Aquatic Diseases

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Abstract

Aquatic environments host a very diverse variety of diseases that may negative impact on aquatic life and humans when transmitted by zoonotic agents. Studying the metabolic and genetic pathways that are responsible for such diseases is important to produce effective mitigation approaches. Because CRISPR-Cas9 technology enables specific genome editing and gene function in marine organisms and the diseases they carry, it opens novel possibilities to investigate zoonotic exposure. This technique makes it easier to find the genetic factor that influences severity, disease hazards, and host-pathogen interaction. CRISPR-Cas9 is a powerful tool to provide a platform for developing fish species that are genetically resistant to diseases and a model to study the dynamics of diseases. Investigators can learn more about the evolution of pathogens and cross-species transmission by examining the genetic cause of zoonotic diseases. Highly efficient sequencing, bioinformatics, and CRISPR-Cas9 combination to speed up the identification of significant zoonotic pathways. The current study illustrates how CRISPR-Cas9 could help identify aquatic diseases, leading the way to novel therapeutic and preventive approaches.

Keywords: CRISPR-Cas9; Zoonotic; Aquaculture; Genomics; Bioinformatics

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Introduction

Zoonoses known as zoonotic diseases are transmitted from animal to human or transmissible diseases that people may acquire from animals. The global aquaculture and fishing industry is facing serious challenges by fish and aquatic-derived zoonosis adverse health hazards (Rahman et al., 2020). Gram-positive bacterial zoonoses such as *Mycobacteriaceae*, *Streptococcaceae*, and *Erysipelothricaceae*; and Gram-negative such as *Aeromonadaceae*, *Vibrionaceae*, *Pseudomonadaceae*, *Enterobacteriaceae*, and *Hafniaceae* are divided into two categories. Zoonotic pathogens also include protozoa like *Cryptosporidium* spp. and parasites like nematodes like *Anisakis* spp.; trematodes like *Opisthorchis* spp. and cestodes like *Diphyllobothrium* spp. Furthermore, people are at threat from fungi associated with sporotrichosis and basidiobolomycosis (Ziarati et al., 2022). Humans acquire the majority of zoonotic diseases by ingesting raw or poorly cooked fish food and fish products. Human exposure to fish zoonosis is very rare but can have serious consequences (WHO, 2021). Recently, discovered that zoonitic diseases are the primary cause of developing infectious diseases influencing golably (Meurens et al., 2021).

CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) and CRISPR-associated (Cas) technology have changed gene editing due to their reliability, efficiency, and cost-effectiveness. CRISPR-Cas9 targets foreign genetic factors such as phage and plasmids and uses RNA-guided endonuclease to cause site-specific breaks in the DNA structure of harmful bacteria. Cas9-DNA linkage and structural changes enable complementary base pairing between the guide RNA and the target DNA which helps in this process. Synthetic single-guide RNA (SgRNA) enhances this RNA-programmable DNA editing method more easily (Jiang & Doudna, 2017).

CRISPR-Cas9 technology offers novel approaches to prevent zoonotic diseases by modifying animals' reservoirs, focusing on disease vectors, cutting the pathogen transmission cycle improving disease surveillance, and promoting vaccine development. It also facilitates the investigation of immunotherapies and possible contenders for traditional therapy. It is a useful tool for combating zoonotic infections because of its accuracy and flexibility. However, challenges like adverse consequences, ethical conflicts, and regulatory complexity need a collaborative effort to ensure beneficial and effective execution (Adnyana et al., 2024).

In this current chapter, we explore the potential of CRISPR-Cas9 in identifying and characterizing genetic factors that contribute to the transmission and virulence of zoonotic aquatic diseases. Additionally, we highlight the applications of CRISPR-Cas9 in developing targeted interventions for controlling and preventing the spread of zoonotic pathogens from aquatic organisms to humans.

2. Background on Zoonotic Aquatic Diseases

The term "zoonoses" is derived from the Greek words "zoon," meaning animal, and "nosos," which means disease. The World Health Organisation (WHO) defines zoonoses as any infection or disease that is naturally transmissible between vertebrate animals and humans, or vice versa. Particularly, approximately 61% of human diseases have a zoonotic origin (Chomel, 2014).

2.1. Effects of Zoonotic Diseases on Public Health

The development and transmission of infectious diseases are greatly influenced by interactions among people, animals, and the environment. Animals are the source of many transmissible diseases that affect humans. According to the 2010 report "Asia Pacific Strategy for Emerging Diseases," over seventy percent of these viruses originate from animal species, and over sixty percent of new human diseases are zoonotic. In recent decades, newly developing diseases in humans have been directly related to animals and animal-derived foods (Thompson, 2019).

2.2. Common Zoonotic Pathogens

Numerous infections may result in zoonotic diseases, which are categorized according to their aetiology. Bacterial zoonoses include diseases such as anthrax, salmonellosis, tuberculosis, Lyme disease, brucellosis, and plague, while viral zoonoses encompass rabies, AIDS, Ebola, and avian influenza. Parasitic zoonoses involve infections like trichinosis, toxoplasmosis, trematodosis, giardiasis, malaria, and echinococcosis. Fungal zoonoses are less common, with ringworm being a notable example. Rickettsial zoonoses include Q fever, whereas chlamydial zoonoses are represented by psittacosis. Mycoplasma zoonoses, such as *Mycoplasma pneumoniae* infection, also pose health risks. Additionally, protozoal zoonoses involve acellular non-viral pathogenic agents, including transmissible spongiform encephalopathies like Mad Cow Disease (Rahman et al., 2020).

2.3. Exposure Route

Humans may contract pathogens from animals either directly or indirectly:

- Direct zoonoses: These occur when diseases are transmitted directly through the air or another medium. A well-known example of a virus spreading by droplets or fomites is avian influenza.
- Animal Bites: The rabies virus, which belongs to the Rhabdoviridae family, is the cause of rabies, one of the deadliest zoonotic diseases. The saliva of a rabid animal, like a dog, bat, or fox, can carry the virus into the human body.
- Vectors: Ticks and mosquitoes are among the vectors that spread diseases like dengue fever. Any animal that may spread a disease can serve as a vector (Huang et al., 2019).

Based on the route of transmission, zoonotic diseases are further divided into:

- Anthropozoonoses: Diseases like rabies that are spread from animals to people.
- Zooanthroponoses: Diseases that humans can spread to animals, such as TB in monkeys and cats.
- Amphizoonoses: Bidirectionally transmissible diseases, such as staphylococcal infections (Rahman et al., 2020).

2.4. Zoonoses in Fish and Aquatic Environments

Several fish-borne zoonotic pathogens have been discovered. Although fish are frequently immune to these diseases, humans can become severely ill as a result. These diseases are often spread through aquatic environments, which are commonly contaminated by residential waste, animal and human waste, or agricultural runoff (Gibello et al., 2016). Humans can get the disease by consuming raw or undercooked fish, as well as carelessly handling aquatic animals or their products. *Aeromonas hydrophila*, *E. coli*, *Yersinia*, *Brucella*, *Shigella*, *Salmonella*, *Streptococcus iniae*, *Clostridium botulinum*, *Klebsiella*, and *Edwardsiella tarda* are some fish pathogens. Vibrio species such as *Vibrio cholerae*, *Vibrio parahaemolyticus*, *Vibrio vulnificus*, and *Vibrio damsela* are among at least 12 Vibrio species that may be zoonotic. When people consume contaminated seafood, these bacteria typically produce severe symptoms such as diarrhea, vomiting, and dehydration (Vega-Lopez, 2020).

Furthermore, fish can spread diseases via wounds or cuts on their skin. Non-tuberculous mycobacterial infections, which are common in aquariums, can cause chronic granulomatous diseases in both humans and fish. In humans, *Mycobacterium marinum* produces skin lesions known as "fish tank granuloma." Fisheries employees who handle live or dead fish are commonly infected with the Gram-positive bacteria *Erysipelothrix rhusiopathiae*, which causes erysipeloid in humans. Symptoms include edema and skin discoloration (Malek et al., 2019). An emerging zoonotic bacteria *Lactococcus garviea* can cause septicemia in fish but in humans, ingestion due to poorly cooked fish and raw seafood spreads endocarditis and encephalopathy. *Nocordia* ssp. are pathogens that cause granulomatous diseases in fish and humans by contaminated wounds. People with compromised immune systems are more vulnerable to nocardiosis which can cause infection in skin pneumonia and ulcers (Meyburgh et al., 2017).

2.5. Impact on Human and Aquatic Health

For the treatment of zoonotic diseases, it is essential to understand the reciprocal link between humans, animals, and the environment. The “One Health Concept” promotes collaboration among sectors such as wildlife biology, veterinary medicine, agriculture, ecology, microbiology, and epidemiology demonstrating holistic approaches. The “One Health Concept” especially in developing nations, helps to alleviate poverty and ensure food and health security by addressing the entire health of humans animals, and ecosystems (Al-Tayib, 2019).

2.6. Organization Collaboration

Cooperation among several sectors is essential for reducing the number of newly emerging reoccurring zoonotic diseases. The European Commission, WHO, OIE, FAO, US Center for Diseases Control and Prevention (CDC), US Department of Agriculture (USDA), and United Nations System Influenza Coordination (UNSIC) all advocate for the execution of comprehensive prevention and control measures guided by One Health Concept (Rahman, 2017).

3. Role of CRISPR-Cas9 in Aquatic Disease Research

3.1. Overview of CRISPR-Cas9

CRISPR/Cas9 is a unique gene-editing technology that has revolutionized the biomedical research field. Proper genome editing is made possible by the fast, effective, and simple modification of genetic errors as well as the synchronization of gene expression in cells and organisms.

CRISPR/Cas9 has a wide range range of scientific applications including the establishment of cellular and animal models functional genomic screening and live genomic imaging (Zhang et al., 2021).

1. This technology has effectively treated genetic mutations or abnormalities in animals by repairing damaged DNA.
2. Human embryos can modified using CRISPR/Cas9 technology.
3. Therapeutic applications include gene therapy, HIV treatment, and personalized materials for cancer and other conditions.

3.2. How Does the CRISPR-Cas System Work?

The two primary components of CRISPR/Cas9 are guide RNA (gRAN) which identifies the target gene and the second is Cas9 protein which acts as endonucleases to produce a double-strand break (DSB) in DNA enabling genome editing shown in (Figure 1 & 2). This technique provides outstanding accuracy and flexibility for genome editing (Ratner et al., 2016; Redman et al., 2016).

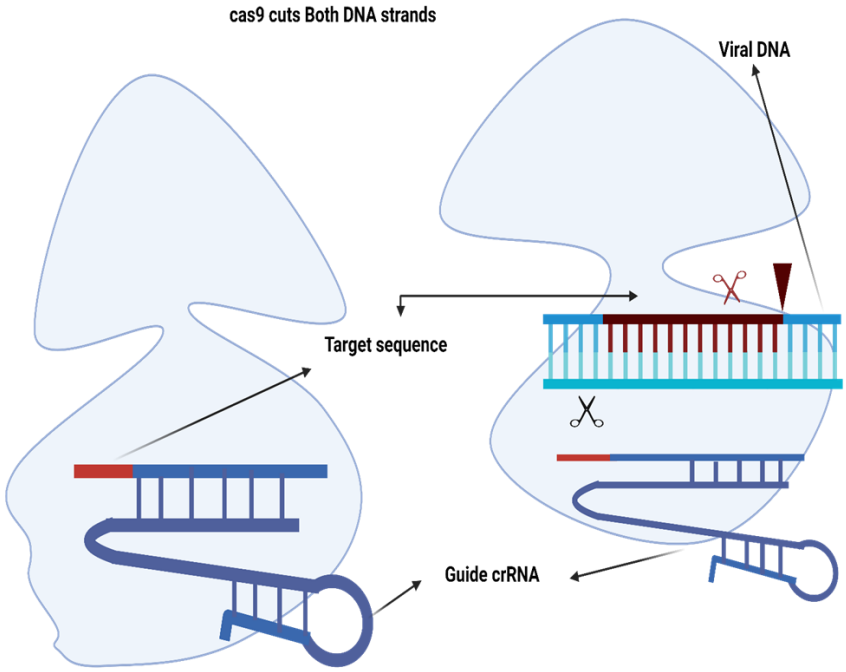

Fig. 1: Cas9 cuts DNA stands as an endonuclease

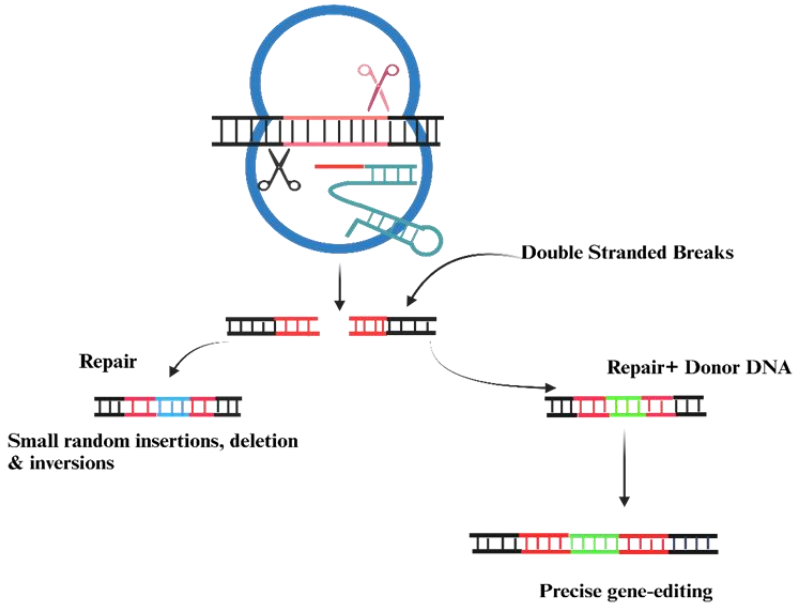

Fig. 2: Gene- editing and repairing.

The three phases of the CRISPR/Cas immune response are adaptation, expression, and interference.

1. **Adaptation Phase**: When Cas protein identifies a particular short DNA sequence called the protospacer adjacent motif (PAM) they attach to the target DNA. The modified complex copies repeat at the 5' end of the CRISPR array after the protospacer has been integrated as a spacer (Figure 3).

2. **Expression Phase**: The spacer sequences and portions of the flanking repeats are present in the mature crRNA that originates from the pre-CRISPR RNA (pre-crRNA).
3. **Interference Phase**: The mature crRNA leads the Cas protein complex to find and split the protospacer sequences in an invading viral or plasmid genome to eliminate the risk (Hille & Charpentier, 2016; Lander et al., 2016).

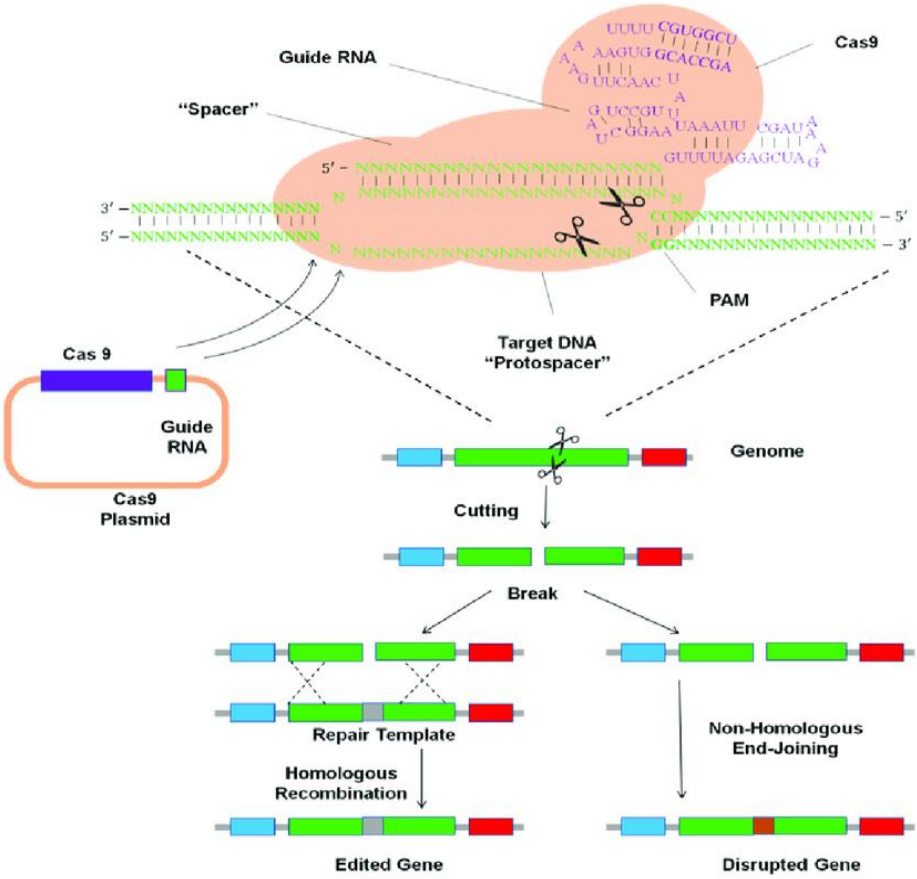

Fig. 3: Complete mechanism of Gene editing via CRISPR technology (Upadhye et al., 2023)(Licence CC-BY 4.0).

3.3. Applications in Host-Pathogen Interactions

Pathogens rely on specific virulence mechanisms to overcome the host immune system and spread diseases that are influenced by the dynamics host and their environmental interaction. Recent techniques such as genome extraction, CRISPR/Cas9, and omics technologies (e.g., dual RNA-seq) have enhanced our knowledge of host-pathogen interaction by revealing the invasion process, immune evasion strategies, and infection-related molecular interactions (Baddal, 2019). Developing effective treatment is essential to understanding the microbe's mutualistic and parasitic relationship with its host. During infections, both the pathogen and the host face significant changes in gene expression with the host usually changing the host gene expression, protein pattern, and epigenomic machinery to survive. Identifying the gene transcript expressed during infection provides important information about pathogenes (Gomez-Diaz et al., 2012).

CRISPR/Cas9 and other molecular technologies enable a specific and simultaneous assessment of the different stages of infection including immune subversion, tissue invasion, and intracellular survival. These outcomes contribute to the formation of medicines that are selective for certain phases of diseases (Ansori et al., 2023). Due to these dynamic changes in host-pathogen interactions, each phase needs to be discussed fully for a better understanding of the same. Prior genomic work in immunobiology offers renewed scopes in infectious diseases and establishes innovative routes to powerful antimicrobial treatments. (De Monerri & Kim, 2014).

3.4. Uses of CRISPR-Cas9 in Aquatic Organisms for Genetic Studies

Numerous types of cell varieties are capable of generating knockout mutations by application of CRISPR/Cas9 (Strich & Chertow 2018). They pointed out that this happens through the use of Cas9 guided by single-guide RNA (gRNA), which results in a DSB that is joining through nonhomologous end joining (NHEJ). This mechanism normally leads to frameshift mutations which produce nonfunctional proteins (Jiang & Doudna 2017). They may create a tool for genome-wide screening to translate various genetic changes with the help of CRISPR/Cas9 technology to study thousands of genetic modifications at once. CRISPR/Cas9 system production has superior on-target efficiency and fewer side effects when compared with RNA interference (RNAi) (Smith et al., 2017).

3.5. CRISPR-Cas9 Used for the Identification of Zoonotic Disease

CRISPR/Cas9 has provided new concepts in terms of detection, and management of zoonotic diseases. It can help change the animal population, identify disease vectors, develop better and more effective disease surveillance, and help in vaccine development. The remarkable

reliability and flexibility of the technology may constitute an effective approach to the control of zoonotic diseases as a result of its ability to change genomes effectively. For instance, possible applications and implementation of the technique can also plant specific genetic components that exist in disease transmission vectors or reservoirs (Adnyana et al., 2024).

4. CRISPR-Cas9 Applications in Illuminating Zoonotic Pathways

4.1. Editing Pathogen Genomes to Understand Virulence and Zoonotic Potential

Genome editing is an emerging technology and CRISPR-cas9 and TALEN make it possible to genetically manipulate aquaculture taxa quickly and effectively. The procedure makes it possible to bring positive changes into the potential genes and enhance the rate of genetic development. Diseases resistance genome editing approaches include the following.

- In selective breeding activities, functional allele segregation is removed or stabilized within the broodstock population.
- Integrating beneficial variations from different populations
- Developing beneficial alleles de novo.

CRISPR/Cas9 has been used in aquaculture for salmonid, Pacific oysters, Nile tilapia, and gilthead sea breams. However, debates continue about whether genome editing constitutes mutation and hence should be closely monitored (Mandrioli, 2022). Diversity in genome editing legislation has contributed to its adoption. Educating stakeholders and the public on the benefits and risks of genome editing is important to worldwide acceptance (Maynard et al., 2021).

4.2. Studying Genetic Mutations That Facilitates Cross-Species Transmission

Public health is seriously threatened by newly developing viral infections, as wildlife hosts are frequently the cause of deadly diseases including human influenza, Ebola fever, and severe acute respiratory syndrome (SARS). These diseases usually develop when animal viruses modify themselves to infect people and then proliferate among human populations. Similarly, viruses can cause the establishment of epizootic diseases when they move across various animal hosts. The recent epidemics of very lethal H5N1 influenza A in birds, which have resulted in hundreds of reported human diseases and fatalities due to "spillover" incidents, are an alarming example. Because of its ability to infect an inclusive variety of avian and mammalian species, its frequent zoonotic transmissions, and its potential to cause human pandemics, influenza A is considered a classic host-switching virus (Wolfe et al., 2007).

Three phases may be distinguished in the viral host switching and disease onset process: Initial Spillover: Often known as a "dead-end" event, a single infection takes place in a new host without any additional transmission. Localized Transmission: Within the new host population, spillover infections start a few short-lived chains of transmission before die out. Stable Transmission: Within the new host population, the virus develops endemic or stable epidemic transmission (Parrish et al., 2008).

4.3. Using CRISPR to Develop Fish Models for Studying Zoonotic Diseases

Many people rely on fisheries and aquaculture for their food and livelihood, and they are essential to the stability of our ecosystem. Aquatic species-related viral diseases have recently resulted in significant financial losses. Developing fast and reliable diagnostic tools is critical for treating patients and preventing the spread of disease. Although modern diagnostics are very sensitive and specific, they are typically expensive, time-consuming, and need specialized personnel, preventing them from being utilized in low-resource settings or for broad screenings during epidemics. CRISPR-based technologies provide deployable diagnostic tools for identifying emerging epidemics, offering a feasible alternative to point-of-care nucleic acid detection strategies. Several CRISPR/Cas technologies such as Cas3, Cas9, Cas12, and Cas13 have been used to create complicated and specific diagnostic tests that leverage techniques such as horizontal flow detection, fluorescent, colorimetric, and signal amplification. These advancements have the potential to change molecular diagnostics, cutting expenses and increasing accessibility. The rising threat of epidemics such as MERS, SARS, and COVID-19 as well as zoonotic diseases, emphasizes the critical need for these technologies. CRISPR-based diagnostic provides an extraordinary opportunity to transfer global healthcare and epidemiological surveillance. (Kostyusheva et al., 2022).

5. Insight into Disease Mechanism

5.1. Unveiling Genetic and Molecular Mechanisms of Zoonotic Disease Transmission

The concept of One Health illustrates how the health of people animals and the environment are interrelated, especially in aquatic environments. Because environmental changes like rising temperatures, fluctuating salinities, and oxygen depletion increase pathogen virulence and stress hosts, marine bivalves are particularly sensitive to climate change. In warm waters e.g., Vibrio species develop more quickly and active pathogenicity genes which raise the risk of zoonotic diseases. Heat stress affects the health of the host and increases the risk of diseases in both animals and humans (Leal et al., 2022). The significance of the integrated method has been demonstrated by research on aquatic zoonotic diseases which have illuminated pathogens epidemiology, host-parasite dynamics, and genetic mechanisms. Global health protection and efficient disease prevention are made possible by expanding One Health knowledge (Antuofermo et al., 2023).

5.2. Genetic Markers for Zoonotic Risk Assessment

Among the various applications of molecular markers in livestock and aquaculture organisms are quantitative trait loci (QTL) strains and hybrid identification, evaluating of genetic variation and diversity, lineage analysis, marker-assessment selection (MAS), and genomic selection (GS). By finding molecular markers linked to QTLgenomic region that include genes that significantly influence particular traits, breeding programs can use MAS to enhance qualities if the markers characterize a significant amount of genetic variation. Furthermore, genomic selection integrates data from hundreds of markers to determine genomic breeding values (GEBVs) in genetic evaluation. This marker-based technique is most beneficial for enhancing characteristics in candidates of selection such as disease characteristics that are difficult or even impossible to essay (Taylor, 2014).

5.3. Stress-Induced Immune Suppression and Zoonosis in Aquatic Organisms

The life cycle of aquatic organisms is full of stress due to climate change like hot and cold temperatures, and food shortage. Cortisol, norepinephrine, and adrenaline stress hormones are produced due to these stress circumstances impacting the immune system, and productivity capability of the animals. As the current research evidence seeks to show, microbes are rather sensitive to stress hormones in a manner that enhances the virulence mechanism for bacterial growth and accelerates the speed of infectious diseases. The information is called on when addressing animals that are derived from stress (Inbaraj et al., 2022).

6. Public Health Implications and Zoonotic Disease Management

CRISPR-Cas9 genome editing technology has made it possible to detect complex diseases at their early stages and enhanced the control of prevention through better diagnostic supports. This study showed the following ways in which CRISPR-Cas9 is helpful in the management of zoonoses such as disease vectors changing animal reservoirs, and the surveillance of diagnostic vaccines, traditional drugs, immunotherapy, and immunosuppressive drugs (Proudfoot et al., 2019).

The impact of CRISPR-Cas9 technology on the global includes changing the phenotypic attribution of mosquitoes, ticks, mites, and arthropod vectors that transmit diseases of humans to animals and the environment (Sylvatic cycle). The technique is designed to improve public health by either minimizing or eradicating these vector capacities to spread diseases by gradually modifying them (Subica, 2023).

6.1. Using CRISPR Data to Enhance Diagnostics

These abilities are particularly useful in environments with a high prevalence of zoonotic diseases and limited laboratory diagnostic resources. Even in isolated areas, CRISPR-based surveillance systems enable rapid detection and response to zoonotic disease outbreaks in a range of places such as clinics, public health labs, field labs, and point-of-care facilities. The One Health approach to observing and controlling zoonotic diseases will move forward if this technology is modified for use in continuous environmental, animal, and human health screening (Ansori et al., 2023).

6.2 CRISPR-Cas9: Transforming Vaccines, Drugs, and Immunotherapy

Vaccines against zoonotic diseases can be developed using CRISPR-Cas9 technology. CRISPR-Cas9 technology can create an effective vaccine by changing the pathogen's DNA to make it less virulent or more immunogenic. CRISPR has enabled the development of new vaccine platforms like viral vector vaccine vaccine and RNA vaccine which can induce robust, powerful, and durable immune responses against zoonotic diseases. These innovations have the potential to reduce vaccine production time, improve effectiveness, and address distribution and production difficulties. The primary synthesis of inactivated viral replicates in vaccines ensures a sufficient vaccine supply. Furthermore, development in immunotherapy models has shown the potential to combat zoonotic disease by targeting and eliminating organisms via immunity (Koshandam et al., 2024).

The development of immunotherapies for zoonotic diseases including most advanced monoclonal antibodies adaptive T-cell therapy and stem cell treatment has been completely transformed by CRISPR- technology. Immunotherapeutic techniques against zoonotic diseases can be enhanced by employing CRISPR to change immune cells to express chimeric antigen receptors (CARs) or enhance antipathogen activation action. CRISPR-mediated genome editing in animal models can facilitate preclinical testing, improving the clinical use of immunotherapies to produce reliable efficient data (Kruglova, 2024).

7. Challenges and Ethical Considerations

7.1. Risks and Challenges of CRISPR Applications in Aquatic Systems

Although CRISPR-Cas9 technology has the potential to significantly decrease zoonotic infection, it comes with drawbacks and moral and ethical challenges. Because accidental changes might impact diseases, hosts or non-targeting species deviation effects are a major cause of stress. The development of an accurate CRISPR system as well as the method of evaluating and reducing these effects are necessary to address this. Furthermore, pathogens may develop strategies to evade targeting the vast genetic diversity of zoonotic diseases and vactor may lead to resistance to CRISPR therapies. The efficiency of these medicines might also be affected by differences in host genetic makeup. Examples of mitigation activities are targeting multiple loci, improving a delivery method technique, and monitoring resistance strains (Caplan et al., 2015).

7.2. Environmental Impact of Genome Editing in Aquatic Species

Controversial issues related to ethical and social implications of changing genetic codes with advanced technology. A potential limitation of CRISPR-Cas9 in its practical application includes positive distribution outcomes and risk, off-target effect, and temporal, spatial, and genotypic effects on diverse biotic systems. The problem arises how to manage GMOs introduced into the environment, how to modify animal physiology, and accidental changes that occur to non-target species concerns ethical issues. An example of using such technology is the case with the implication of Wolbachia in Indonesia to cure and prevent dengue disease. These are complex ethical and social issues and therefore critical discussion with multiple stakeholders, including researchers, academics, policymakers, epidemiologists, ethnicities, topical medicine, public health providers, virologists, environmentalists, and the community must be encouraged (de Graeff et al., 2019).

8. Future Directions

8.1. Combining CRISPR-Cas9 with Complementary Strategies

Therefore, to assess the applicability of CRISPR-Cas9 technology in zoonotic diseases therapeutic in the future, a multidimensional perspective is needed. The Indonesian government can leverage the CRISPR-Cas9 genome editing technology to improve global health and well-being and avoid zoonotic diseases. Current and future efforts in improving CRISPR intervention targeting zoonotic diseases are under the

following categories: 1) developing a new delivery system for CRISPR; 2) fine-tuning CRISPR; 3) exploring the usage of new gene drive system; 4) embracing the state of art engineering approaches. Moreover, partnerships between government and non-profit organizations business partners, academic institutions, and the affected communities are also required for the development of CRISPR intervention (Brokowski, 2019).

8.2. Expanding Research on Aquatic Species and Pathogens

Interdisciplinary cooperation stimulates the application of research outcomes into useful suggestions regarding social programs by sharing information resources and knowledge. Safe and ethical use of CRISPR-Cas9 technology and zoonotic disease prevention and control activities demand an innovative and permissive regulatory system that minimizes technology transition. Ideally, empowerment efforts should target educational researchers, technical zoonotic staff, physicians, legislators, and members of the community at moderate or low risk with CRISPR-Cas9 (Gostimskaya, 2022).

Conclusion

In conclusion, the CRISPR-Cas9 technology recommended a favorable position to support the exploration of the genetic aspects of the zoonotic relation in aquatic diseases and provide knowledge of pathogen-host interactions and disease transmission routes. It may contribute to the determination of the resistance mechanism and virulence factors because it enables the correct error of the genome, which may be revealed in new methods of control. This technology enhances understanding of aquatic diseases and their epidemics at the same time exploits health by reducing opportunities for zoonotic threats. The synchronization of CRISPR-Cas9 with other advanced forms of genetic engineering promises to revolutionize disease diagnosis and treatment in aquaculture. Besides, the developing disease-resistant aquaculture species have a significant implication for the sustainability of the aquaculture industry. To realize its entire effectiveness and possibilities distinctive lines of work must be united as well as more research made.

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