

Human Journals Review Article

November 2019 Vol.:14, Issue:1 © All rights are reserved by Mahnoor Patel et al.

Development in Fish Vaccinology



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Submission:	27 October 2019	
Accepted:	02 November 2019	
Published:	30 November 2019	





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Keywords: Adjuvants, Delivery Methods, DNA Vaccine, Fish Vaccination, Inactivated Vaccine

ABSTRACT

Sustainable development in aquaculture is equivalent to disease prevention, and vaccination has become the single most important tool. There has been a dramatic reduction in the use of antibiotics since the introduction of oil-based vaccines. Fish can be vaccinated by immersion and the oral route. However, the protection falls short compared to the injection of a single dose of naked DNA into the fish muscle. Nevertheless, the prospect of having a commercial product on the market within 5 years in meagre. New technologies are promising but it is more likely there will be improvements of existing vaccines than completely new technologies taking over the fish vaccination scene in the next 5-10 years.

1: INTRODUCTION

Sustainable development of aquaculture relies on disease prevention. Vaccines stimulate the immune system to help fight off diseases and the application of these methods to control infectious diseases are growing in importance. Perfecting the use of adjuvants and delivery systems is needed to meet the demand for vaccines to ensure the safe supply of healthy fish products [1-3]. Because of the increase in infectious diseases there is an insightful and reliable positive attitude for vaccination to prevent the diseases at the early level [6]. Vaccines are generally use for the stimulation of an immune system for the protection against particular diseases. Like mammals, fish also have an immune system for the protection against diseases, also helps them for the existence and maintenance of their integrity in any kind of unfriendly environment [7]. Vaccines which are developed for use in aquaculture is able to reduce the use of an antibiotics which are important in fish production. Next generation vaccines which are based on multiple killed antigens delivered in combination with adjuvants for enhancement in vaccine effectiveness [8-10].

2: Vaccination of Fishes:

Vaccines stimulate the adaptive immune system to mount a response against a pathogen or rather against defined structures of the pathogen, the immunogenic parts. Vaccination has been used as a prophylactic means for decades and it has been estimated that ten percent of all cultured aquatic animals are lost because of infectious diseases alone, the vaccines are either delivered by an intra-peritoneal injections, by immersion, where animals are placed in a vaccine solution, or by oral administration [2-5]. Live modified vaccine having live pathogens which has been reduced non-pathogenic or avirulent with the help of physical, chemical or genetic engineering. Live vaccines which are modified typically maintains their ability for infecting the host that can allow for operative presentation of protective antigens for generating cellular immunity that is CD4 and CD8 T-cell response of cell mediated immunity. Modified live vaccines have an advantage for easily delivered with the help of immersion to young fish for the stimulation of both humoral and cellular immunity for the long duration. There are few disadvantages which includes some issues with the live modified vaccines safety to host and the environment. A successful modified live vaccine which can use in warm water aquaculture is used for highlighting the live vaccine strategy [6].

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3: Vaccination Strategies:

The choice of delivery method or combinations thereof is of crucial importance for obtaining good protection. Further to this, if there is a need to protect the fish at an early stage of the life cycle, immunocompetence has to be considered and, in general, the recommendation would be to wait for the fish to reach an age where it can mount an appropriate immune response. The assumption is that to protect fish before the development of full immunocompetence, one would have to rely on innate immunity or innate immune responses since it is known that responses mature before the animal being fully immunocompetent (*i.e.* being able to respond by an adaptive immune response) [2-4].

4: Adjuvants and Principle of Action:

The mode of action of adjuvants, in general, is poorly understood. It is known that the formation of a depot at the site of inoculation is a typical trait of many of the adjuvants resulting in slow release of the antigens and the presentation of an antigen to immunocompetent cells. This is a typical feature of oil-based adjuvants and most likely plays a key role in the induction of immunity for many of the fish vaccines currently available for use in different aquaculture markets [2, 3].

5: Vaccination Injection:

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5.1: DNA Vaccine:

Gene therapy can be defined as the delivery of a therapeutic gene for expression in somatic tissue. There has been a rapid development in the field of gene therapy and DNA vaccination since the expression of foreign genes *in-vivo* was demonstrated. Subsequently, it was known that the injection of naked plasmids into the muscle could also elicit an immune response. DNA vaccines do not need the gene to be permanently expressed as transient expression of the gene is sufficient for evoking the immune response [3].

It has been demonstrated that DNA vaccination induces a strong and protective immunity to some viral infections in fish, the challenge, as regards DNA vaccination, is that so far, with a few exceptions, induction of protective immunity has been reliant on intramuscular injection. Immersion is a delivery route offering many advantages compared to conventional ways of administration. Use of cationic liposomes as a delivery system for DNA by the immersion

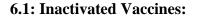
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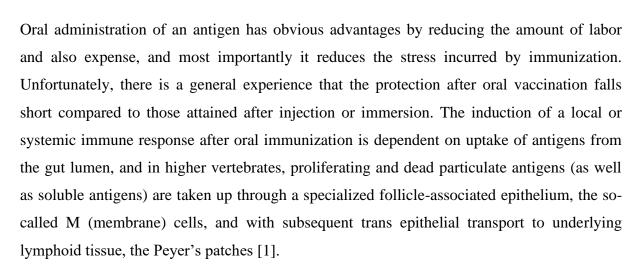
route has met with severe toxicity problems. The mechanism of the acute toxicity is suggested to be an interaction between the cationic liposomes and anionic components of gill mucin. The consequence is hypoxia and this is most likely the cause of acute toxicity observed in rainbow trout fry [2].

The safety of DNA vaccines for use in fish is more of a concern that their efficacy. Safety issues are related to integration into chromosomal DNA, pathological processes at the site of injection, distribution to internal organs and longevity of retention of foreign DNA in these organs. Issues related to tumorigenicity will probability raise public concern and potentially also with the regulatory bodies [4].

It has been demonstrated that retention and expression of antigens at the injection site appear for an extended period. However, not beyond 4-5 weeks post-vaccination. The local reactions at the site of injection are prominent and last for an extended period and much longer than the actual antigen expression, as detected by immunohistochemistry. Strong inflammation, muscle cell destruction and granuloma formations are evident at 3 and 12 weeks postvaccination [5].

6: Oral Delivery:





Despite the observation that vaccine efficacy in fish is so limited after oral delivery, there are very few studies that address the uptake and transepithelial transport in enterocytes of soluble versus particulate antigens. The morphological or functional characterization of enterocytes is also scant, yet there are indications for a regional specialization of the gut epithelium

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concerning the uptake of macromolecules, and the hindgut enterocytes are considered important in this respect [2, 4].

6.1.2: Vector Vaccine:

A new principle has also been explored whereby genetically modified *E. coli* expressing the exotoxin A of *Pseudomonas aeruginosa* have been fed to live Artemia brine shrimps. The Artemia has been subsequently fed to zebrafish [3, 5].

Route of Administration	Type of Formulation/Delivery Methods	Advantages	Disadvantages
Injection	 Oil-based (water- in-oil, oil-in-water or w/o/w) Liposomes (experimental) 	 Most potent with little waste of vaccine Allows the use of adjuvants Cost-effective method for high- value species Mass vaccination is possible 	 Stressful Impractical for fish >15g Labor Intensive Injection-site reactions Immune response (level of protection)
Immersion (inactivated and live vaccines)	 Used to a limited extent (mainly in marine fish species) Live attenuated vaccines Vector vaccines 	 Large scale application Moderate stress to the fish Easy-allows mass vaccination of immune-competent fish High efficiency using live, attenuated vaccines 	 A large amount of vaccine is needed Low efficiency for inactivated vaccine Inferior to injection routes in terms of efficacy Cost prohibitive for large fish
Oral delivery	 Top-dressing The formulation in PLG (experimental) 	 Imposes no stress on the fish Moderate cost All fish sizes can be vaccinated when immuno- competent Usually safe- primes mucosal immunity (external surfaces) 	 Usually low efficacy Can be cost- prohibitive for larger fish

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