[Article ID : 01/I/15/0121]

DNA VACCINE IN FISHES

[1]Anurag Semwal, [2]Mahendra Kumar Yadav

[12]Ph.D Scholar at College of Fisheries,
G.B. Pant University of Agriculture and Technology
Pantnagar, Uttarakhand

Introduction

Fish disease is one of the most important consequential factors in aquaculture sector. When disease outbreaks occur, diagnostics are conducted to detect the cause, and then treatment recommended to fish like oral treatment, an immersion (a dip or a bath), or, in rare cases, an injection treatment. Expenses come from delayed production, treatment chemicals, mortalities, and labor can be significant. Two techniques of disease prevention that have been commonly utilized in other animal industries are immunostimulants and vaccines. A vaccine is any biologically based preparation proposed to build up or to improve immunity to a specific disease or group of diseases. A vaccine, if effective, can help prevent a future loss from being a major economic drain. There are limited numbers of DNA vaccine strategies that have been efficient in giving significant protection against fish diseases. The great exemptions are DNA immunization against viral hemorrhagic septicemia virus (VHSV) at experimental level and infectious hematopoietic necrosis virus (IHNV) at commercial level. According to the Norwegian Biotechnology Advisory Board DNA vaccination is "the intentional transfer of genetic material (DNA or RNA) to somatic cells for the purpose of influencing the immune system".

What should be the properties of the ideal vaccine?

1. It should be safe for the fish, the person(s) vaccinating the fish, and the consumer
2. It should protect against a broad strain or pathogen type and gives 100% protection
3. It should provide long-lasting protection, at least as long as the production cycle
4. It should be easily applied
5. It should be effective in a number of fish species
6. It should be cost effective and
7. It should be readily licensed and registered

Evolution of DNA vaccines in Aquaculture

In 1996, the first DNA vaccination of fish took place, when Anderson and his coworker vaccinated rainbow trout (*Onchorhyncus mykiss*) against infectious hematopoietic necrosis virus (IHNV). After that, several trials are performed for a good sort of fish species and pathogens. In 1999 the injection of Atlantic salmon with pCMV4-G (plasmid-encoded glycoprotein) from a rainbow trout IHNV isolate induced outstanding defense against challenges with IHNV, albeit the salmon were much larger than the rainbow trout in earlier studies. Similar to what is observed in mammalian species, DNA vaccination of fish has been shown to induce adaptive and innate immune responses and seems particularly productive against novirhabdoviruses (like IHNV and VHSV). These are simple RNA viruses with five or six genes and a single protein of the viral surface (glycoprotein, or G protein) serving as a protective antigen. An immunization against VHSV in rainbow trout allows the stimulation of cell-mediated immune responses involving both CTLs and natural killer (NK) cells and has also been shown to significantly reduce the replication of virus during challenge. In 2005, a vaccine against IHNV infection in salmonids was also one among the primary DNA vaccine ever to

be cleared for marketing (by the Canadian Food Inspection Agency). During 2017, a polyprotein-encoding DNA (CLYNAV (Elanco), vaccine against Salmon Pancreas Disease Virus infection in Atlantic salmon used within the European union, based on a positive risk benefit assessment following analysis of data. It was approved for use in Norway by the Norwegian Medicines Agency.

Recent DNA vaccination laboratory trials

Effects of DNA vaccines against different viral and bacterial disease in fish have been observed and reviewed by Tonheim et al., Kurath, Redding and Weiner, and Gomez-Casado et al. Seemingly, DNA vaccination may also give protection against bacteria and parasites but not against all.

Pathogen	Gene inserted	Host	Administration route/adjuvant
IHNV	IHNV-G plus suicidal gene	Rainbow trout	Intramuscular/none
IHNV	IHNV-G; different genogroups	Rainbow trout	Intramuscular /none
IHNV	IHNV-G	Rainbow trout	Oral/PLGA
VHSV	E. tarda as delivery vehicle of the vaccine	Olive/Japanese flounder (Paralichthys olivaceus)	Intramuscular
IPNV	VP2; Segment A of TA strain	Atlantic salmon	Intramuscular
Megalocytivirus	86-residue VP	Turbot (Scophthalmus maximus)	Intramuscular
E. tarda	D15-like surface antigen	Japanese flounder	Intramuscular
V. harveyi	DegQ or/and Vhp1	Japanese flounder	Intramuscular
Flavobacterium psychrophilum	Hsp60, hsp70	Rainbow trout	Intramuscular
Cryptobia salmocitica	Metalloprotease	Atlantic salmon and rainbow trout	Intramuscular

Table 1. Experimental DNA vaccines in fish following experimental infection

Administration of DNA vaccines

Intramuscular injection is commonly used in fish for the delivery of pDNA and typically results in clear transgene expressions at the injection site. This initial dispersion of a vaccine can be sufficient for very small fish to ensure the perfusion of intact pDNA into more distant tissues, while the injected volume would predominantly rest along the needle trajectory in large fish. Intravenous, intraperitoneal, oral delivery and particle bombardment are other routes of pDNA administration that have been investigated in fish.

Advantages, disadvantages and challenges of DNA vaccines

When provided at early life stages, DNA vaccines demonstrate high efficiency and have the advantage of inducing protective immunity across a wide temperature spectrum. The benefits of DNA vaccination continue to grow beyond mere immunological abilities. When looking at the concept from the point of view of a producer or/and investor, DNA vaccines are relatively cheap and simple to produce. For all DNA vaccines, the processes needed for development are similar,

and the simplicity of cloning also allows for rapid modifications in a way that is not normally obtainable with traditional vaccine preparations. Potential side effects include, for example, the possibility of autoimmunity, immune tolerance to the expressed antigen, excessive CTL response leading to myositis, chromosomal integration, inflammation of the injection site and loss of tissue.

Safety and regulatory aspects by DNA vaccines

Safety aspects include potential impacts on the vaccinated animals, consumer and the environment. Other safety concerns include the possible shedding of the vaccine from vaccinated animals and predatory animals into the environment. Human protection also involves possible consequences from self-injection by vaccinators. When protection aspects need to be reported, these safety aspects need to be taken into account by the appropriate authorities.

Conclusions

There is a critical need to increase the efficacy of DNA vaccines against recurrent and difficult-to-combat viral infections, which can be met by: (i) The use of vaccine carriers to increase the absorption of antigen presenting cells accompanied by increased transgene peptide antigen presentation, (ii) Use of nano-scale particles to increase the degree of cross-representation of such cells may also be helpful in generating antibody response and immunity mediated by cells, (iii) Using additional adjuvants such as TLR ligands, other than RNA and/or DNA, to substantially improve the response. Protection and regulatory uncertainties are connected to the distribution and degradation of DNA after injection, and it is important to make further effort to understand the processes of pDNA uptake, from the moment of administration to the stage of transcription and translation in the nucleus.

References

Adomako M, St-Hilaire S, Zheng Y, Eley J, Marcum RD, Sealey W, Donahower BC, LaPatra S, Sheridan PP: Oral DNA vaccination of rainbow trout, Oncorhynchus mykiss (Walbaum), against infectious haematopoietic necrosis virus using PLGA [Poly(D, L-Lactic-Co-Glycolic Acid)] nanoparticles. J Fish Dis 2012, 35:203–214.

Anderson ED, Mourich DV, Fahrenkrug SC, LaPatra S, Shepherd J, Leong JAC: Genetic immunization of rainbow trout (Onchorhynchus mykiss) against infectious hematopoietic necrosis virus. Mol Mar Biol Biotechnol 1996, 5:114–122.

Board TNBA: Regulation of DNA vaccines and gene therapy on animals.2003.

Corbeil S, LaPatra SE, Anderson ED, Kurath G: Nanogram quantities of a DNA vaccine protect rainbow trout fry against heterologous strains of infectious hematopoietic necrosis virus. Vaccine 2000, 18:2817–2824.

Evensen Ø, Leong JA: DNA vaccines against viral diseases of farmed fish. Fish Shellfish Immun 2013, 35:1751–1758.

Gillund F, Tonheim T, Seternes T, Dalmo RA, Myhr AI: DNA vaccination in aquaculture –Expert judgements of impact on environment and fish health. Aquaculture 2008, 284:25–34.

Gomez-Casado E, Estepa A, Coll JM: A comparative review on European-farmed finfish RNA viruses and their vaccines. Vaccine 2011, 29:2657–2671.

Gomez-Chiarri M, Livingston SK, Muro-Cacho C, Sanders S, Levine RP: Introduction of foreign genes into the tissue of live fish by direct injection and particle bombardment. Dis Aquat Organ 1996, 27:5–12.

Grisez, L., and Z. Tan. 2005. Vaccine development for Asian aquaculture. In *Diseases in Asian Aquaculture*, edited by P. Walker, R. Lester, and M.G. Bondad-Reantaso, V: 483–494. Manila: Fish Health Section, Asian Fisheries Society.

Hansen E, Fernandes K, Goldspink G, Butterworth P, Umeda PK, Chang KC: Strong expression of foreign genes following direct injection into fish muscle. FEBS Lett 1991, 290:73–76.

Heppell J, Lorenzen N, Armstrong NK, Wu T, Lorenzen E, Einer-Jensen K,Schorr J, Davis HL: Development of DNA vaccines for fish: vector design, intramuscular injection and antigen expression using viral haemorrhagic septicaemia virus genes as model. Fish Shellfish Immunol 1998, 8:271–286.

Kurath G: Biotechnology and DNA vaccines for aquatic animals. Rev Sci Tech 2008, 27:175–196.

Lorenzen E, Einer-Jensen K, Rasmussen JS, Kjaer TE, Collet B, Secombes CJ, Lorenzen N: The protective mechanisms induced by a fish rhabdovirus DNA vaccine depend on temperature. Vaccine 2009, 27:3870–3880.

Lorenzen N, LaPatra SE: DNA vaccines for aquacultured fish. Rev Sci Tech 2005, 24:201–213.

Myhr A, Dalmo RA: Introduction of genetic engineering in aquaculture: Ecological and ethical implications for science and governance. Aquaculture 2005, 250:542–554.

Purcell MK, Laing KJ, Winton JR: Immunity to fish rhabdoviruses. Viruses 2012, 4:140–166.

Purcell MK, Nichols KM, Winton JR, Kurath G, Thorgaard GH, Wheeler P, Hansen JD, Herwig RP, Park LK: Comprehensive gene expression profiling following DNA vaccination of rainbow trout against infectious hematopoietic necrosis virus. Mol Immunol 2006, 43:2089–2106.

Redding L, Weiner DB: DNA vaccines in veterinary use. Expert Rev Vaccines 2009, 8:1251–1276.

Tonheim TC, Bøgwald J, Dalmo RA: What happens to the DNA vaccine in fish? A review of current knowledge. Fish Shellfish Immunol 2008, 25:1–18.

Traxler GS, Anderson E, LaPatra SE, Richard J, Shewmaker B, Kurath G: Naked DNA vaccination of Atlantic salmon Salmo salar against IHNV. Dis Aquat Organ 1999, 38:183–190.