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Health & Welfare
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Development of edible vaccines for aquaculture



1 October 2000 Joe Jilka, Ph.D.



Feed-based medicines could reduce labor costs and stress



Although still in the development stages, corn-based feed with built-in vaccines may soon be available to the aquaculture industry.

With many aquafeed ingredients becoming more expensive and/or harder to find, grain-based feeds – particularly those based on corn – are receiving more attention. Now scientists are developing vaccines for the animal healthcare market that can be delivered directly through corn grain. By coupling the vaccines with conventional feed formulations, vaccines can be customdelivered with no more effort than feeding fish at customary levels.

Transgenic plants and protein expression

With improved technology in the past decade, transgenic plants have been successfully used to express a variety of proteins. In 1999, over 60 million acres of transgenic plants were grown, indicating that the system of production of transgenic plants is stable and robust.

Many additional proteins have been expressed in plants solely for their antigenic potential, including viral proteins and subunits of bacterial toxins. Animal and human immunization studies have demonstrated the effectiveness of many of these plant-derived recombinant antigens in stimulating the immune system. In fact, ProdiGene, a biotechnology company based in Texas, USA, recently demonstrated a corn vaccine for swine that, when fed to the swine, protected them from infection by the transmissible gastroenteritis virus.

Benefits for vaccine production

The utilization of transgenic plants for vaccine production has a number of potential benefits over traditional vaccines in aquaculture disease management:

- Direct feeding is possible, since the vaccines are expressed in corn, significantly reducing labor costs and stress on the fish.
- Since there are no known human or animal diseases that are able to infect plants, concerns with contamination are eliminated.
- Vaccine production in transgenic crops relies on the same technologies established to sow, harvest, store, transport, and process the corn as those already used for feed crops, making corn a very economical means of large-scale vaccine production.
- Expression of the vaccine in the seed, which is the plant's natural protein-storage compartment, maximizes the stability of the vaccine. This in turn minimizes the need for refrigeration and keeps transportation and storage costs low.
- Multivalent vaccines are possible by either blending the seed of transgenic corn lines containing other vaccines into a single multivalent vaccine, or expressing multiple antigens in the same plant. These edible vaccines could be used in a stand-alone vaccination strategy, as a booster, or in combination with other vaccines and vaccination routes.

Mucosal immune response

One of the most promising aspects of edible vaccines is the ability of orally administered antigens to stimulate a mucosal immune response. Mucosal surfaces – the linings of the respiratory, gastrointestinal, and urogenital tracts – play an important physical and chemical role in protecting animals' bodies from invading pathogens and harmful molecules. Since most invading pathogens first encounter one or more of the mucosal surfaces, stimulation of the mucosal immune system is often the best first defense against many transmissible diseases.

Development of edible vaccines

ProdiGene is in the process of developing a number of edible vaccines in both the human and animal health areas. In particular, corn expressing viral proteins of swine transmissible gastroenteritis, an important swine disease, is currently being used in swine feeding trials. Preliminary data indicate that these vaccines protect the animals from viral infection.

Hybrid seed production

Once an inbred seed line is developed that contains the vaccine of interest, hybrid seed is produced that incorporates the necessary agronomic qualities needed for the area of production. ProdiGene has a partnership with Stauffer Seed of Aurora, Nebraska, USA, which produces the seed for ProdiGene and implements the grain production program.

Stauffer also performs other tasks, such as warehousing, inventory, and delivery of seed to the contract grower. It also implements grower contracts that specify production practices and quality-control measures, and provides agronomic support to producers for maximizing the yield and quality of the grain.

Optimizing production

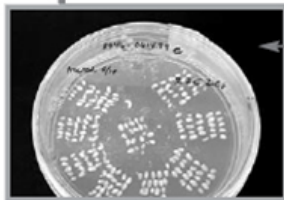
ProdiGene and Stauffer work together to plan grain production by selecting the best growing areas, and identifying farmers capable of producing high-quality grain and ensuring identity preservation of the grain. Proper measures for isolation from neighboring crops are specified and documented. Approved production practices, including pesticide and fertilizer applications, are recommended and documented.

Trained agronomists work with the growers to help with best management practices and scheduling of grain delivery. At harvest, equipment and storage facilities are cleaned and the grain is assigned lot identification numbers for tracking through processing. Production records and management practices are recorded for future documentation.

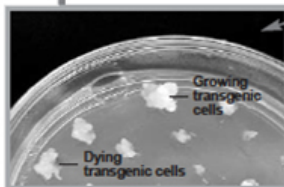
Regulatory compliance

Grain processing and seed fractionation are currently done as toll processing at existing mills. Good Manufacturing Practices (GMP) are used in compliance with the established regulatory guidelines for processing identity- preserved grains and ProdiGene's own quality assurance guidelines. The GMP practices include cleaning equipment with compressed air or vacuum, weighing and analyzing the incoming bulk material and existing fractions, labeling, inspecting the integrity of packaging, and preparing procedures for storage and shipment of fractions to another site for further processing if needed. Additional processing may involve oil removal, protein extraction, protein purification, or other operations.

How to Make a Transgenic Corn Plant



1. Immature embryos are harvested from pollinated corn and incubated with bacteria engineered to carry the gene of interest. The bacteria transfers the gene into some of the cells in the immature embryos.



2. Plant hormones are added, which causes the embryo cells to turn into a mass of cells with no form or function called callus tissue. The plate also contains an herbicide. Since the bacteria was engineered to transfer a gene for herbicide resistance along with the gene of interest, the cells altered by the bacteria can grow in the presence of the herbicide, while all other cells are killed.



3. The callus tissue that now contains only transgenic cells is put on a new plate. There, plant hormones cause the tissue to form roots, stems, and leaves in the presence of light. All previous steps were kept in the dark.



4. The tiny corn plants are carefully picked off the plates and planted in test tubes to allow them to grow. This step ensures the plants are strong enough to survive in the greenhouse.



5. Six to nine months later, transgenic seed is available in the greenhouse. This seed is screened to make sure the level of transgenic protein is high enough for the intended purposes. Note the paper bags that ensure the pollination process is completely controlled.

Conclusion

Edible vaccines for aquaculture will be easy to administer by simply incorporating the expressed vaccine in grain in feed. These vaccines have the potential to eliminate much of the labor involved in current aquaculture health management strategies, reduce the stress of vaccination on fish, and open up new strategies for aquaculture health management.

(Editor's Note: This article was originally published in the October 2000 print edition of the Global Aquaculture Advocate.)

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