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From Labs to Libraries: The Significance of Cell Line Depositories

M. Yaswanth Babu[1], J. Joselin Karunya Mary[2], B. Tejaswi[3] and H. Siva kumar[4]

[1]M.F.Sc. Fish Biotechnology Research Scholar, Central Institute of Fisheries Education, Panch Marg, Off. Yari Road, Versova, Andheri (west), Mumbai, Maharashtra, India

[2]M.F.Sc. Fish Biotechnology Research Scholar, Central Institute of Fisheries Education, Panch Marg, Off. Yari Road, Versova, Andheri (west), Mumbai, Maharashtra, India

[3]M.F.Sc. Fish Biotechnology Research Scholar, Central Institute of Fisheries Education, Panch Marg, Off. Yari Road, Versova, Andheri (west), Mumbai, Maharashtra, India

[4]M.F.Sc. Post Harvest Technology Research Scholar, Central Institute of Fisheries Education, Panch Marg, Off. Yari Road, Versova, Andheri (west), Mumbai, Maharashtra, India

SUMMARY

An essential biological tool for doing research in the domains of physiology, virology, pharmacology, toxicology, cancer/carcinogenesis, and transgenics is provided by "cell lines." A variety of tissues, including the ovary, fin, swim bladder, heart, spleen, liver, eye muscle, vertebrae, brain, and skin, have been used to create "fish cell lines." Another area that shows promise for using cell culture to boost agricultural output is cellular agriculture. In vitro models are currently being studied for cell-based meat manufacturing, which might alleviate strain on open-water fishing and establish a secure and long-lasting food supply. Additionally, the increased use and advancement of cell lines pave the way for the establishment of cell line depositories, where cells can be placed, kept safe, and made accessible to researchers for additional study.

INTRODUCTION

Food availability in the appropriate quantity for all the people is a global challenge that never goes away. Food and nutritional security are major worldwide concerns now because of the strain on resources, a deteriorating climate, and an expanding population. Fish is regarded as "**nature's superfood**" among other foods since it is a great source of calcium, vitamin D, iodine, good fats (such as omega-3 fatty acids), proteins, and other nutrients. In comparison to the worldwide consumption of 20.5 kg in 2018, India's per capita fish consumption in 2019–20 was 5–6 kg. The amount of fish consumed per person has been rising steadily over the last few decades, and this trend is predicted to continue in the years to come. According to FAO, the global demand for fish and fish products is growing. Utilizing the variety of blue foods and utilizing cutting-edge technologies like cellular agriculture, we need to keep up with the latest advances to fulfill the increasing demand for blue foods.

Cell line characterization:

Fish cell line development has produced an exponential increase in cell lines, which highlights the need for precise characterization and verification of fish cell lines to provide repeatable scientific results. The extensive instructions for employing cell lines address several topics, including misidentification, contamination with bacteria, and features of cell culture, and provide suggestions for resolving these concerns.

1) Identity testing
- DNA Barcoding
- Chromosome analysis

2) Purity testing
- Sterility testing for bacterial contamination
- Mycoplasma testing
- Transmission electron microscopy (TEM)
- PCR analysis for specific viruses

3) Immunocytochemistry for cell morphology
4) Growth studies
5) Transfection efficiency

Cell line depository:

The growing interest in creating fish cell lines, of which 918 have been reported globally, is indicative of the growing use of cell lines in such promising areas. 280 fresh fish cell lines from 44 fish families across 21 nations were created in just the last ten years. The development of these cell lines is only as vital as their upkeep and long-term storage. By allowing other researchers to get and use a cell line, the developer of the line receives acknowledgment and avoids duplication of effort when it is deposited in a depository. These depositories are crucial for the storage and preservation of genetic material, but they can also serve as effective tools for conservation in particular situations, such as the extinction of a species as a result of overfishing, natural disasters, or other issues. Popular and reliable cell line depositories include the German Collection of Microorganisms and Cell Cultures (DSMZ), the American Type Culture Collection (ATCC), and the European Collections of Cell Cultures (ECACC). It has been noted that the vast majority of fish cell lines found in these depositories are from temperate freshwater species that are significant to the European and American economies. The production of cell lines from Asia has increased due to substantial studies in fisheries and aquaculture in Asian nations such as China, India, Vietnam, Thailand, etc. Cell line depositories were established as a result of the growing requirement to meet the demand for Asian fish species cell lines. With funding from the Department of Biotechnology, Ministry of Science and Technology, Government of India, New Delhi, a depository known as the National Repository of Fish Cell Lines (NRFC) was founded in 2010 at the ICAR National Bureau of Fish Genetics Research (NBFGR), Lucknow, India, to serve as a platform for the deposit of fish cell lines developed in the country. Developing, depositing, maintaining, and distributing verified fish cell lines upon request is the mission of NRFC.

Cell line applications:

Pathogens primarily bacteria, fungi, and viruses found in aquatic environments operate as limiting factors, causing illnesses and impeding fish production in freshwater, brackish water, and marine habitats. Fish diseases, particularly viral infections, have become more common as intensive aquaculture systems have grown, and transboundary movements of valuable ornamental fish have increased fish disease frequency. Fish cell lines can be used to accurately identify and isolate these diseases, as well as evaluate appropriate chemicals or medications for their elimination.

Disease monitoring:

Fish viruses such as Nodavirus (NV), nervous necrosis virus (NNV), infectious pancreatic necrosis virus (IPN), cyprinid herpes viral hematopoietic necrosis virus (HVHNV), tilapia lake virus (TiLV), red seabream iridovirus (RSIV), white spot syndrome virus (WSSV), etc. have been tested for their susceptibility in numerous fish cell lines. Fish cell lines have been used to identify and isolate viruses, enabling screening affected individuals and developing techniques to lower infection and increase aquaculture output. Another example is the first-ever identification and isolation of the fish nodavirus in India, which causes significant mortality in sea bass hatcheries. Aside from viruses, fish cell culture has also been utilized to identify bacterial pathogens that are severely harming fish production, such as *Vibrio anguillarum, Estrella lausannensis, Parachlamydia acanthamoebae, Photobacterium damselae, Damselae Piscirickettsia, Renibacterium subsps. Salmonis Salmoninarum, Yersinia ruckeri* etc.

Environmental monitoring:

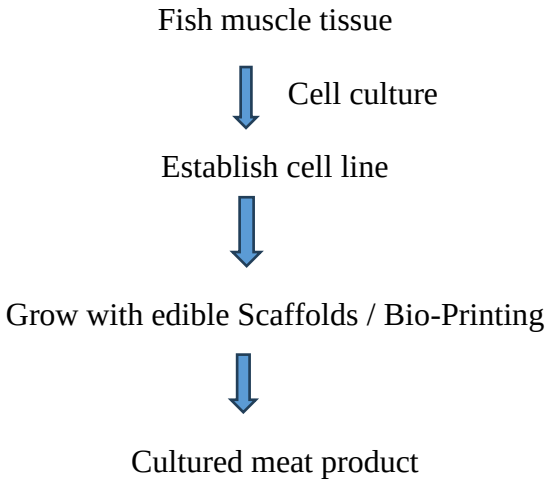
Artificial inputs have been used more frequently in the new aquaculture systems. Farmers with issues with their fish's health have resorted to using pesticides and medications to regulate their ponds. When conducting ecotoxicology investigations, such as evaluating the cytotoxicity and estrogenic activity of chemicals and environmental materials, primary cell cultures and cell lines might be useful instruments. Control over the macro- and micro-environments, including pH, osmolarity, temperature, and nutritional components, is one of the major benefits of employing cell lines as an in vitro model. A cell line generates a larger quantity of cells that are better suited for low-volume replication and long-term storage with good recovery rates. Drugs and other chemicals are frequently evaluated for safety and potential mechanisms of action through studies on cells derived from living organisms. Researchers have used cell culture methods to understand the underlying processes of the immunomodulatory effects of heavy metals, pesticides, fungal toxins, etc., and their links with fish health. Comparing the relative cellular susceptibility of different species to different environmental toxins, such as

pesticides, heavy metals, and other aquatic pollutants from agricultural runoffs, is also made possible by the widespread use of fish cell lines. Such data will be useful for monitoring aquatic habitats and for classifying contaminants according to their cytotoxic effects. Recombinant fish cell lines could strengthen their usefulness as a bioanalytical instrument for evaluating the environment.

Cellular agriculture:

Cellular agriculture is one of the exciting and possible technologies to boost the production of certain agricultural commodities. To create certain agricultural products in vitro that would otherwise be collected naturally, a combination of molecular and synthetic biology approaches is applied along with cell culture equipment. Since Dr. Mark Post of Maastricht University in the Netherlands presented the first cultured meat prototype in 2013, the cultured meat industry has advanced significantly. According to recent studies, the number of businesses offering cultured meat is rising. Approximately 99 firms globally have been reported as of the beginning of 2023, compared to just 4 in 2016. These companies work on various elements of generating cultured meat. Additionally, it is projected that by 2040, grocery stores, retail establishments, restaurants, and other food outlets will sell around 60% of the meat that comes from culture-based meat sectors globally. This cellular method would also assist in lessening the fishing load on specific fish stocks by facilitating a safe and sustainable food sector. The potential of technology is an exciting prospect and a developing solution to support the growing population, as well as to address the difficulties of sustainable environmental growth and animal welfare related to intensive aquaculture systems. Cell-based fish meat processes may also be used for species such as mahi-mahi and crabs that have a marketable demand but are either costly to harvest or are not farmed.

Procedure of cultured / lab meat production:



Transgenic studies and reproductive biotechnology:

Progress in cell culture techniques has made gene targeting and gene transfer easier for the generation of transgenic fish. Transgenic lines in model organisms or other animal systems have been made possible by transgenic zebrafish created using primary cultures of genetically modified zebrafish male germ cells. A transgenic trout (*Oncorhynchus mykiss*) with a construct encoding the GFP reporter gene driven by a fast myosin light chain 2 (MlC2f) promoter was used to create a genetically modified myogenic cell culture. Primordial germ cell (PGC) cultures can be used to develop the transgenic line. The Vasa marker makes it easier to isolate and characterize individual PGCs in fish for germline-specific expression. Tanaka and colleagues created a transgenic medaka line by expressing GFP in its germ cells. Fish germ cell transplantation proved to be a successful method for producing surrogate broodstock in aquaculture systems. For the first time, rainbow trout seedlings were produced via intraperitoneal transplanting of PGCs. The development of stem cell culture and its subsequent applications in aquaculture biotechnology and in vitro fundamental research will revolutionize the fisheries industry and help bring about the "blue revolution." Transplanting spermatogonial stem cells presents numerous opportunities for an aquaculture species' successful captive breeding program. The nuclear transfer method, in conjunction with PGCs and ES cells, makes the transformation effective for the creation of transgenic fish.

CONCLUSION

To meet the nutritional needs and ensure food security for every member of the expanding global population, new and inventive technologies are always required. One such method to boost agricultural commodity production is cellular agriculture. Fish cell lines are valuable biological resources that can enhance food and nutritional security. They have numerous uses in vital fields, including disease diagnostics, toxicity research, genetic material conservation, and lab-grown meat. In these cutting-edge fields, it is anticipated that the application of fish cell lines will grow in the upcoming years. Depositories of cell lines, such as NRFC, would be useful in the screening process for infections, particularly those that come from fish crossing borders. Numerous fish cell lines are available at NRFC, which gives Indian researchers more opportunities to use them for studies in toxicology, genetics, cytology, and disease management. They can also investigate the development of cell-based meat to expand the food basket.

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