A holistic review on live transportation of food fishes: Current Scenario and Future Prospects

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Abstract

Live fish has emerged as a high demanded commodity in recent past on account of the progressive quality concepts of seafood consumers. Live fish transportation is dependent on several internal as well as external factors which need to be considered critically for improving survival as well as quality during their transportation. The lack of a systematic approach for live fish transportation, beginning with on farm handling till marketing, is the most significant issue faced by the stakeholders. This review provides an integrated insight on the current state of knowledge in the field of live transportation of food fishes emphasizing the significance, current status, challenges and exploration possibilities.

Key words: Live transportation, anaesthetization, stress, water quality, aquaculture, seafood

Introduction

Focused research for the development of value-added products has been rapidly expanding in the recent past so as to meet the expectations of globalized consumers in the market and this trend has also impacted the retail marketing of aquatic produces. Market studies have indicated a good appetite for high-end aquatic products with high quality. Live fishes are ideal sources representing top-end value-added products because they guarantee freshness and have higher price realisation than fresh, chilled, or frozen goods. Marketing live fish attracts consumers for its quality as well as ensuring better revenue for farmers, thereby equalising demand at both ends. Due to developments in logistics, live fish is occupying a specialised segment in both domestic and international markets. Globally live fish trade is well established mainly in most of the South-east Asian and Southern pacific regions[1]. Initially, in India, the domestic market for live fish transportation was confined to the North-Eastern states but the rising demand for these categories paved way for a promising market throughout the Country. However, meeting the demand for the same was challenging and possible only with a few species of carps as well as air breathing hardy fishes like cat fishes are transported[2]. Practices of live seafood transportation protocol extend from use of primitive traditional techniques to huge insulated or non-insulated containers for short distance transportation. For distant trade, sophisticated hauling trucks with filtration, aeration, refrigeration and water reticulation facilities by employing external sources are used[1]. Regardless of the market potential the challenges like stress and associated decrease in survival rate of fishes during transportation limits the exploration possibilities[(2,3,4)].

There are several internal as well as external factors which need to be considered critically for improving survival as well as quality of fishes during their transportation. Density of fish transported as well as water quality parameters like water temperature, pH, dissolved oxygen, carbon dioxide, and ammonia etc. are major determinants[5]. Appropriate designing of container for transportation and adherence to standardized operational procedures in every stage is critical for achieving better survival and quality in live fish transportation. The article attempts to give an overview of live transportation of food fishes and factor involved in it. It is also highlighted that there are a very few comprehensive reviews are available in this area and hence this paper offers updated information for future research in the field of live fish transportation.

Live fish transportation systems

Generally, there are three basic transport systems for live fish trade viz. closed system, open or tank method and the modified waterless system[6]. The closed system is a sealed unit, poised with all the live transportation requirements (Figure 1) whereas open system comprises of water filled containers in which facilities for transportation are supplied from outside[7] (Figure 2). The modified waterless system is operated without any water, except for being stored damped by the usage of pre-chilled materials such as sawdust or cotton thereby maintaining cool and moist conditions [(8)] (Figure 3). Various research have been carried out and patented in the field of live fish transportation including systems for transportation[(9,10,11,12,13)] and method of live fish transportation[14]. Container designing for Container designing for live fish transportation should consider the quantity and quality of fish to be transported by avoiding sharp projections and abrasive internal surfaces[15].

The open transportation system is found ideal for short distances and time duration not longer than two hours. During transport, air or oxygen should be supplied constantly or intermittently and water replacement should be followed at regular intervals of 4-5 hours or less to avoid warming up of water[16]. However, this type of methods involves risk of mortality due to splashing of water and the approach is often constrained by distance and time. Australian seafood industry basically employs three live transportation systems including the polystyrene containers, the pickle barrel system and the big box system[6]. The first system, which is the widely adopted one, employs standard polystyrene box with inner plastic bag and foam liners. The second one utilizes screw top plastic containers whereas the third system employs larger boxes with oxygen cylinders strapped alongside, on top or inside the box lid. Zhang and Lv[17] refined a refrigeration and oxygenation measurement and control device for live fish transportation. The mechanism also had an independent diesel engine driving power that was independent of the vehicle's regulation, and completely free from the impact of traffic and a variety of emergencies, sustaining the living conditions for live fish. A mobile platform based on the requirements related to behaviour, security, availability, operation and remote control of the system was proposed by Curiel *et al*.[18]. The platform aided in real-time monitoring of the water quality during the transportation of live tilapia fingerlings for increasing the survival rate and reducing the stress during transportation. Rifat *et al*.[19] fabricated a simple and energy efficient aerator-cum-oxygen accumulator for live fish transport using electric power from the vehicle used for transportation. An aerator was constructed using a bilge pump (DC) having a capacity of 1100 GPH operated under 12-volt and 3-ampere. The equipment was designed to utilize power from the vehicle dynamo while the vehicle was under operation whereas under static condition the power supply unit switched to the battery of the vehicle. A review of recommendations for live transportation of food fishes is presented in Table 1.

The present technological advancements in these strategies demands water which would likely increase the energy consumption and transportation cost[20]. An alternative approach in live fish transportation for increasing the biomass and survival rate is waterless transport. Authors like Wang *et al*.[21] evaluated the survivability, physiological responses and flesh quality of Amur sturgeon (*Acipenser schrenckii*) during waterless transport. In the study, cold anesthetized fishes were packed into plastic bags which were filled with pure oxygen, sealed and placed on the trays line by line which were further piled up in a refrigerated carriage and the temperature maintained around 4°C. They also suggested packing to be done with minimum stress and injury to the subjects and temperature to be maintained below 15°C.

Factors influencing live fish transportation and associated stress

The survival and quality of live transported animal is the major concern and are dependent on various factors, or combination of them. It is these factors (Figure 4) that affect the pattern of stress response of transported animals, which plays a vital role in the overall survival as well as their quality. Stress in fish manifests as a series of complex physical and physiological changes resulting in its health deterioration. Failure to reduce fish stress during transportation leads to increased incidence of disease and decreased survival[7]. Stress in fish during live during live transportation is attributed by a complex of factors viz. purging, harvesting methods, handling, water quality parameters such as dissolved oxygen levels, increase in carbon dioxide, drop in pH, and rise ammonia levels in the transportation system, biomass in transportation system, transportation time etc.[22] The resistance to physiological changes creates stressors to pile up which leads to significant quality changes. Eventually, it leads to decrease in market value as the ultimate market strategy is to provide healthy fish that survive until they are sold and processed or after they are re-stocked[23]. Therefore, exporters are cautious not only in mitigating the loss of product due to mortality, but also to minimise the product quality deterioration on account of shipping stress[24]. Stress includes evident physical symptoms like colour variations[25], speedy respiration[26], behavioural changes[27] while delicate, invisible effects include variations in the fish blood which drastically reduce its capacity to abide variations in water quality[28]. Countering pH reduction could be the prime factor during short transportations of less than eight hours duration whereas in long transportation involving more than eight hours, ammonia accumulation is a major concern[29]. Plasma cortisol is the most often used stress marker, and it is acutely elevated during brief episodes of transportation but stays elevated even during long-transport cases[30]. Potential stress factors for live fishes in waterless transportation are ambient temperature, ambient relative humidity and levels of oxygen and carbon oxide[31]. Indication of primary stress response of juvenile cod due to handling events during transportation were analysed by invasive measurements of free cortisol release into the tank water by the subjects[4]. Wang *et al*.[21] in their studies reported blood glucose and serum cortisol as reliable markers of stress meanwhile alanine aminotransferase and lactic dehydrogenase were the significant markers that reflected the extent of physiological effects on fish caused by stress responses. Anaesthetics were found useful for calming excitable fishes thereby reducing injuries whereas it could also bring forth a biological response similar to that caused by stress[32]. Adding sodium chloride to live transport tanks was found helpful to minimize the effects of transport stress. Biswal *et al*.[33] evaluated the stress alleviating effect of NaCl during simulated live transportation of *L. rohita* fingerlings using various biomarkers approaches. They also suggest the addition of 0.4% NaCl in the water for mitigating long-term transformational stress and reducing mortality. According to studies, the enzymes such as serum glutamic pyruvic transaminase[34], lactate dehydrogenase[8], malate dehydrogenase[35] and G-6 phosphatase[36], the stress hormone cortisol[37], metabolites like triglyceride[38] and creatinine[34], and expression of HSP70 mRNA[39] could be successfully deployed as biomarkers to evaluate the degree of transportation stress.

Pre-transportation conditioning

Effective live fish marketing relies to a great extent on the status of the fish prior to live fish transportation. The operational protocol used when transporting live fish includes the handling and treatments given to fish prior to transportation. The quality of the fish will have an impact on its transportation stress and related survival. Fish must be healthy and have sound physical conditions as in contrary, excessive mortalities may happen during or post transportation. Weakened fish is less competent to survive the stress during transportation[22]. Studies also recommend the necessity of having information regarding the health status of fish prior to transportation in order to adopt mitigation measures including lowering of fish biomass being transported, adoption of less stressful harvesting, avoidance of sick and injured fish and careful handling [5, 7].

Right from the harvest, responsible capture and transfer strategies with minimal or no injuries and stress should be adopted for effective transportation protocols[40]. Certain recommendations to minimize the effect of transport on fish welfare have been laid out by OIE Aquatic Animal Health Code, 2019[41] which describes the individual duties and responsibilities involved in fish handling throughout transportation. It is recommended that harvesting be done early in the morning or at night when the ambient temperature is lower so as to avoid drastic temperature variations[42]. Less stressful harvesting techniques like line fishing, trapping or harvesting using knotless meshed nets are best recommended for fin fishes. Further, the basic prerequisite for a stress-free environment is the assurance of sufficient clean and oxygenated water for the harvested ones[41]. The captured fish in highly stressed condition will have elevated respiration rates and may cause secretions like mucus as well as excreta, which in turn affect the water quality of the holding tank. Studies have shown that keeping fish under starvation, referred to as purging, for one to two days would be advantageous to achieve better survival rates [21, 42]. During starvation, the gastrointestinal tract is completely emptied which prevents excess secretion of faeces, reducing the bacterial decomposition as well as ammonia accumulation and reduces the oxygen requirement ensuring acceptable water quality during the transport[22]. Further, fish with empty stomachs are hardier as the energy spent for digesting the food can be used to adapt to a stressful environment.

Anaesthetization

Prime objective of live fish transportation is to supply the commodity to the destination with maximum possible survival which can be realized by maintaining the fish under minimal stress. This can be achieved to a greater extend by reducing the metabolic rate of the fish during transportation. Extended transportation at low mortality rate can be achieved by lowering the respiration as well as metabolic rate through anaesthetisation[43]. There are several anaesthetisation techniques such as use of anaesthetics, application of low temperature and carbon dioxide. Anaesthetics can be categorized into natural and synthetic ones. Several synthetic agents employed for anaesthetization include tricaine methane sulfonate (MS-222)[44], phenoxy ethanol[45] and etomidate[46]. However, accumulation of residues in the meat and adoption of withdrawal period before harvesting limits the usage of synthetic anaesthetics[47]. Various plant based natural compounds such as eugenol, menthol, globulol, linalool, guaiol, dehydrofukinone, cineole, spathulenol, caryophyllene oxide, carvacrol, thymol and myrcene have been studied to have anesthetization effects on fishes[47]. However, most of these natural compounds are not listed under generally recognised as safe (GRAS) and can be lethal to fish as well as create risk to consumers, limiting their application. Temperature-induced cold anaesthetization can be considered as an alternative to the usage of anaesthetics for live transport technologies[48] Fundamental theory underlying temperature-induced anaesthetization is reducing the temperature of the water to a bearable minimum of the subject thus tranquilizing or immobilizing them[49]. Lowering the temperature during transport sedates the fish, lowers their metabolic rate and increases the oxygen saturation level[42]. Generally, temperature reduction is used to sedate the fish rather than absolute anaesthetisation wherein the subjects exhibit weak opercular movements and retain equilibrium to a greater extent[49].

Water Quality

During live fish transportation, the variations in water quality parameters are complex and inter-related. An increase in fish density thereof can exacerbate the water quality issues. Water quality in closed fish transportation systems is a function of loading density and the transportation duration. The main changes in water quality during live transport are low dissolved oxygen levels, increase in carbon dioxide, drop in pH, and rise ammonia levels. These changes are identified as potential limiting factors, owing to excessive respiration and carbon dioxide and ammonia excretion by transported fishes[3]. A time-series experiment showed that most water quality degradation occurs rapidly, within the first hour after packing. Fishes are prone to chronic stress when exposed to poor water quality, improper stocking densities and inadequate diets for a prolonged period[41]. Water temperature is considered as a critical parameter during live fish transportation as it influences and induces rapid variations in other quality parameters such as pH, dissolved oxygen, dissolved carbon dioxide, ammonia etc. Rate of water exchange and stocking density are the determining factors that affects water quality in open systems whereas decrease in pH, accumulation of toxic metabolites such as carbon dioxide, ammonia, organic carbon leads to deterioration of water quality in closed systems of live fish transportation[50]. Studies have indicated that oxygen carrying capacity of transporting water could be raised by lowering water temperature[50]. Svobodova *et al*.[51] recommended minimum dissolved oxygen requirements as 6 ppm (70% saturation) for cold water fishes, 5 ppm (80% saturation) for tropical freshwater fish and 5 ppm (75% saturation) for tropical marine fish. During live fish transportation, dissolved oxygen should be maintained around 100% saturation or carbon dioxide level should be kept below 20 ppm. Toxicity of ammonia indicates direct relation with pH and reported lethality even at low level (2 ppm) which could be minimized by fasting fish prior to transport and or adding ammonia-reducing agents to the transport water[52]. During simulated live transportation of cod (*Gadus morhua*)[4], marked drop in pH within an hour was observed, which further lowered below 7 after 6 hours of transportation. Buffers such as TRIS, sodium bicarbonate and magnaspheres were found advantageous for maintaining pH during live fish transportation. The acceptable pH for fish normally ranged between 7.5 - 8.5 and was recommended to maintain total alkalinity values above 50 ppm[53]. Low pH results in physiological problems primarily affecting the oxygen utilizing ability leading to respiratory stress even when dissolved oxygen levels are high. This declining pH in fish transport applications is the direct effect of carbon dioxide accumulation[54]. A variety of chemical packs that produce oxygen[55] as well as ammonia/carbon dioxide scavengers for live fish transport applications are employed viz., direct immersion packs, packs placed indirectly outside the inner bags for release of oxygen by fine airstone inside the bags[56]. Prototype live fish transport systems using sodalime were found effective in reducing carbon dioxide levels and increasing pH in the transport medium[6].

Revitalization process and post transportation survival

Transferring transported fish to holding tanks at their destination is a simple process, but it must be done carefully to avoid fish mortality. The water quality in transport bags will be considerably different from that in the holding tank, and the fish should be transferred with extreme caution[57]. The survival rate associated with the transported fish is highly influenced by the conditions used during revitalization. Immediate aeration of the transported environment is necessary to remove accumulated carbon dioxide and clean saltwater equal to the volume in the transported box was added to acclimatize fish to holding system conditions[6]. An increased survival rates were obtained for fishes when allowed to recover in saline enriched water after live transport[58]. Addition of sodium chloride in the transport water was found to be advantageous to improve survival rates.

Status and prospects of live fish trade

Live fish trade has been increased in the recent past on account of the lucrative market for these commodities. Live, fresh or chilled fish represents the largest share (44 %) of fish utilized for direct human consumption as being often the most preferred and highly priced form of fish[60]. South East Asia is the dominant market for the live fish and Hong Kong is considered as the centre of live food fish trade. A well-established supply chain starting from fishermen to the end consumers had been developed in coastal waters of South East Asia. In the Indian context, live fish products are sold on a modest scale to both domestic and international markets. It creates additional revenues for aqua farmers and generates considerable foreign exchange for India by opening up new markets for seafood exports. The country has exported 7287 MT of live fish worth Rs.324.26 Crore during 2019-20, which is a high price realization compared to the other processed commodities like chilled forms[8]. Currently, the major live items exported from India is mainly crustaceans including mud crab, blood clams, horn shell, murex, lobsters etc. with the major export hub of India being Chennai and market being South East Asian countries. However, a few challenges associated in this live export trade include the higher air freight charges, delay or logistic interruptions resulting in higher mortality, bulk and cost of live transportation systems and lack of well-established supply chain. Even though a few live transportation systems are commercially available, most of the stakeholders are not willing to adopt such technologies on being unaffordable. On considering practical feasibility, simple transportation systems like Styrofoam boxes are used as alternative, especially for waterless transportation purpose but they pose adverse environmental consequences.

In the present scenario, the live fish trade urges for a smart live fish transportation system self-capable of manipulating the transport conditions to accommodate a wide range of live species. The various water additives such as antifoaming agents, NH_3 and CO_2 absorbers, other herbal and synthetic aesthetics etc. for mitigating potential stressors during live seafood trade are underutilized on a commercial basis. Despite the widespread application of food traceability system for various seafood, its adoption as well as standard operating procedures (SOP) to be followed in live fish transportation have to be fully developed. Traceability platform in combination with species specific SOP plans would provide optimal micro environmental conditions for the target species with improved survival and quality, minimizing the transportation risks. The introduction of real time quality control mechanisms can dynamically guarantee the survival rate and food quality after development of species-specific optimized protocols. Hence, focused research and development on these aspects can bring about an effective live fish transportation device with due consideration on its economic feasibility so as to meet the needs of stakeholders across the seafood value chain.

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Table 1 Recommendations for live transportation of food fishes

Species	Pre-transportation	Transportation containers	Transport water	Transportation density	Post transportation	Reference
Fresh water aquaculture species in Bangladesh	Purging for 2 days Minimum conditioning period: 12 h Minimum DO level: 4ppm Stocking density: 17.7 kg m^{-3}	Plastic tanks of 1 m^3 capacity; Aeration by using 4-6 hp diesel pump/oxygen cylinder/ battery operated agitators or compressors	Temperature: 25 ºC Dissolved oxygen: 6 ppm	200 kg m^{-3}	Partial exchange of water in transportation tank with stocking tank in order to balance the physicochemical quality of the water	[52]

Tiger Grouper (*Epinephelus fuscoguttatus*)	Cold anaesthetisation from room temperature to 15ºC at the rate of 2ºC/h	Plastic bags filled with oxygen and water	Temperature: 15ºC Ascorbic acid: 25 ppm ß -1,3-glucan: 3.2 ppm	1000 g L^{-1}	-	59
Atlantic salmon (*Salmo salar*)	No purging	40-m live haul vessel having two identical live-holds amidships of 325 m^3 capacity.	-	95.5 kg m^{-3}	-	61
Olive flounder (*Paralichthys olivaceus*)	Purging for one day	Flow-through tank	Natural conditions of temperature and photoperiod	16.5 kg m^{-3}	Post transportation duration: 24 hours	62

Blue Tilapia (*Oreochromis Aureus*)	Acclimatised in closed fiberglass tanks with re-circulated water for two days	Plastic bags filled with oxygen and water	Clove oil: 0.2 ppm	80 g L^{-1}	-	63
Silver catfish (*Rhamdia quelen*)	Purging was avoided to simulate market practice	Plastic bags filled with oxygen and water	Eugenol at the rate of 3.0 μL L^{-1}	169.2 g L^{-1}	-	64
European eel (*Anguilla anguilla*)	Purging for four days	Transported in polystyrene transport tank (0.95x1.05x1 m) filled with 150 L water with recirculation system and supply of pure oxygen	-	270-290 kg m^{-3}	Post-transport stocking density: 72 kg m^{-3} Not fed during the recovery period to avoid adverse water	65

					conditions	
Grass carp, (*Ctenopharyngodon idella*)	Acclimatized and purged for one week with a continuous flow of aerated water	-	Eugenol:10 ppm		-	[66]
Nile tilapia (*Oreochromis niloticus*)	Purging for one day	Fiber glass box of 1 m³ capacity, equipped with diffusers and oxygen cylinder	Sodium chloride: 6 ppt	400 kg m^{-3}	-	[67]
Black Rockfish (*Sebastes schlegeli*)	Purging for three days	Transportation unit consist of seawater circulation pump, ultraviolet sterilizer,	Temperature: 8ºC	198 kg m^{-3}	-	[68]

		oxygen generator, protein skimmer and sea water cooling device Illumination intensity: 60 Lux				
Orange-spotted grouper (*Epinephelus coioides*)	Purging for 3 days	Plastic bags filled with oxygen and water	*Glycine tomentella* extract: 250ppm	200 g L^{-1}	-	[69]

Figure 1 Schematic representation of open system for live fish transportation (a) Container (b) Oxygen diffuser (c) External oxygen source

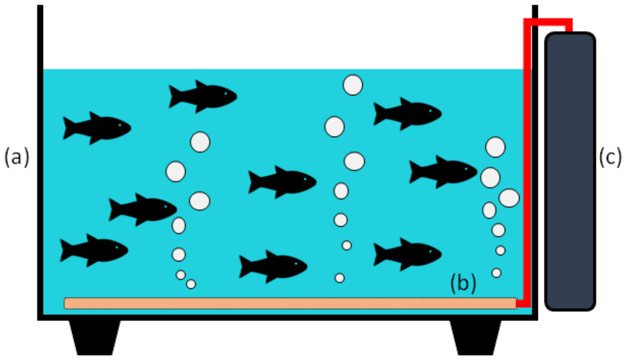

Figure 2 Schematic representation of closed system for live fish transportation (a) Container (b) Pump (c) Filtration unit (d) lid (e) Oxygen diffuser (e) Inbuilt oxygen source

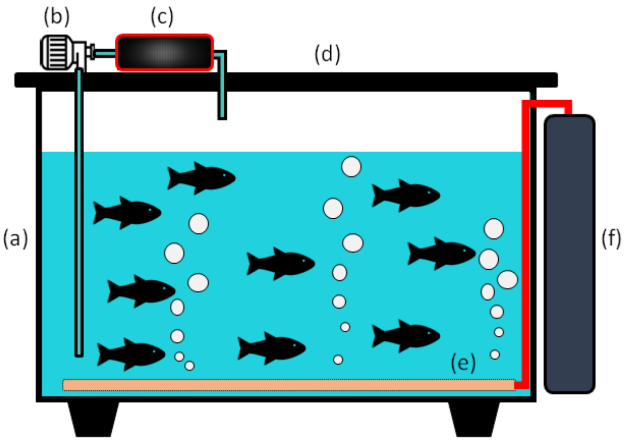

Figure 3 Schematic representation of waterless system for live fish transportation (a) Insulated container with lid (b) cooling unit (c) pre-chilled layering medium (d) live fish/shellfish

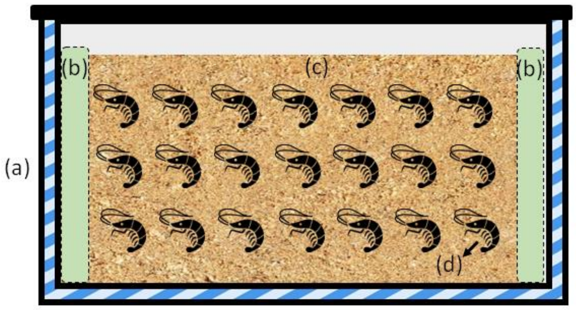

Figure 4 Factors Affecting Live Fish Transportation