

International Journal of Theoretical & Applied Sciences, 14(1): 38-47(2022)

ISSN No. (Print): 0975-1718 ISSN No. (Online): 2249-3247

Meta-analysis of PIT tagging effects on fish growth and mortality

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ABSTRACT: Portable antennas have grown in popularity as a means of tracking fish using Passive Integrated Transponder (PIT) tags. PIT tags have made it possible to conduct scientific work into a variety of ecological features of animals since they were first developed in the middle of the 1980s for a range of uses including fisheries and aquaculture. The small, alphanumeric coded chip injected into individual animals provides more precise measurements of growth rates, feeding and breeding behaviours, movement patterns, and survival rates than the conventional methods of outwardly marking animals for identification. Fish ecology research frequently uses PIT tags, and their viability in migratory species has been thoroughly examined. Animals taken from the wild without permission, including zoo animals, pets, migrating birds, and endangered species, have also had their identities verified using PIT tags. PIT tags are enabling improvements in conservation biology and physiology as well as greater comprehension of the social connections among animals. PIT tags bring up a world of possibilities for resolving intractable animal problems despite their drawbacks, which include a high price, a limited detection range, and the potential for tag loss during migration in some circumstances. There is still a lack of knowledge about the tagging study on the identification of suitable release sites and facilities for continuous stock monitoring. This review on tagging study helps to overcome the seed production of commercially important species such as selective breeding programmes, is required.

Keywords: PIT tag, antennas, breeding, movement, survival, feeding, migratory.

INTRODUCTION

In biological research, PIT tagging is a technique for locating and tracking species. It entails implanting a PIT tag scanner-readable electronic microchip with an alphanumeric code into a person of interest [31]. Since their development in the middle of the 1980s, PIT tags have been widely used for individualised fish monitoring throughout the world, and their popularity has grown [31]. PIT-tags use straightforward tagging procedures and are inexpensive, lightweight, and durable [63]. PIT tags have been used in several animal models since the 1980s to gather biological and population demographic information [31]. Since their introduction in the early 1980s, internal tags, particularly PIT tags, have seen a substantial increase in popularity. PIT tags have been used to identify hundreds of thousands of individual fish. PIT tags have been employed in research on predation rates [9], individual movement [12], feeding behaviour [9], and habitat utilisation [8]. The detection range of a PIT tag is especially important in applications where fish are not recaptured but are tracked remotely using a fixed or

portable antenna. PIT-tag detection data is frequently used to identify stocks [38], track their movements [53], track their migration patterns [41], determine their abundance [2], track their growth, and estimate their mortality [41]. These data cannot be altered by the PIT tags either directly or indirectly. Individual tagging is a typical scientific practise in fisheries. Important ecological and demographic data, such as information on survival [50], development [35], migration [65], and habitat use, can be obtained by identifying and following specific individuals over time and space [47]. PIT tags are frequently used when a large number of fish need to be marked for ecological conclusions because of their low cost and relative simplicity of use [4]. PIT tags are used in numerous large-scale population tracking and activity studies [52]. Fish field research has traditionally included individual animal identification using internal and external tags. External tags such as Jumbo Roto tags, Petersen disc tags and dart tags have been utilised in shark tagging research [43].

There are other tagging techniques, such as painted labels, dart tags, or leg bands, but the bulk of these are exterior, making their codes more subject to environmental factors that could make them difficult to read [62]. PIT tagging, on the other hand, avoids codes from becoming unclear or lost as an internal type of labelling, making it suitable for use in both short-term and long-term research [36, 37]. Brewer et al., [14] revealed that PIT tagging had great retention and survival rates, making it ideal for application [7]. PIT tagging is also been used in reptile experiments [16]. Much research concerning fish physiology and behaviour, such as swimming efficiency, has been used for tagging [28]. On the other hand, these studies have discovered that the PIT tag's implantation and existence had no impact on the measurements of the variable.

Fish must be classified separately since performance varies substantially among individuals of various sizes [3, 41]. This should ideally begin as early as possible and continue for as long as possible. The best

candidates for small fish labelling are PITs [58]. These tiny tags have billions of distinct codes and an infinite lifespan, allowing them to be used on massive fish samples [12, 58]. PIT tags have mostly been used in the management of large fish husbandry [38]. The detection range of larger tags is often wider than that of smaller tags. Small tags can be used on relatively juvenile fish and have less effect on the growth, survival, and behaviour of the animals; but, depending on the species, they typically have a limited detection range.

The effects of the tagging process and the physical effects of tags on animal performance and health have been known to fisheries biologists [39]. Feasibility studies on tag assessment are highly encouraged when there is no detailed data on the individual species available, both to confirm findings and for ethical reasons, as physiological and behavioural responses to specific tagging procedures differ significantly between species [6, 64].



Fig. 1. Tagging using the injector.



Fig. 2. Tag position.

The duration and intensity of the perturbation caused by the tagging protocols and the presence of tag were characterised based on survival, growth, tag retention, healing progress, and body weights to assess the suitability of the tagging technique and to determine the minimum size at which fishes can be successfully tagged. PIT tags have been used for both identifying specific broodstock [38], and for studying individual development, action, mobility, and passage past dams [12, 60]. Using a hypodermic needle to insert a PIT tag into the peritoneal cavity (Fig. 1). has shown to be a very successful technique for tagging juvenile salmonids [58]. Researchers found that the survival of juvenile Oreochromis niloticus (also called Tilapia nilotica) that had been PIT-tagged in this manner was low (10-50%) and positively associated with fish size. This was due to the difficulty of controlling needle penetration [6]. Although this has not been proven for salmonids [58], hypodermic needles used are also been found not sufficient for PIT-tagging warm-water fishes in habitats with fish infections due to poorly tagged fish survival [7].

To PIT-tag juvenile salmonids in the peritoneal cavity, surgical treatments (such as employing scalpels to create incisions for tag insertion) provide an alternative to using a hypodermic needle [6]. Similar techniques were used in the past to implant PIT tags in the body cavity of small *Salmo salar* [60]. The juvenile fish size prevents surgically inserted PIT tags on juvenile salmonids to be verified for retention and survival [60]. Salmonids have long been the focus of tagging efforts due to their ecological and cultural significance.

A. Tag Position

The brood fish's physiology is unaffected by the tag because it is inserted in their dorsal muscles. It makes up a relatively little portion of their total body weight. The majority of the experiments have been done to assess whether it is feasible to implant PIT tags into the body cavity of small juveniles [13, 58]. Body cavity insertions have been performed just posterior to the pelvic fins [6] or just before the fins [33]. PIT tags are frequently inserted into fish by making an incision with a scalpel and injecting them with a needle into the coelomic cavity [7, 58]; or the dorsal muscle [26] (Fig. 2). Forceps were used to manually insert a 125 kHz PIT tag (EM4102 Injectable Transponder Animal Tags; 8.0 2.0 mm, 0.06 g) into the abdominal cavity [4]. An index calibrated using histology data was used to visually monitor the incision's closure [54] (Fig. 4).

Scalpel incisions were used to make an opening through which PIT tags were inserted into the abdomen of *Hybognathus amarus*, a small-bodied minnow. This



Fig. 3. Tag using manual injectors.

B. Post Tagging Retention

PIT tags that are implanted into the peritoneal cavity have demonstrated high retention rates of above 95% in numerous studies [58]. For the majority of field experiments, PIT tag retention (Fig. 5). should be examined to assure accurate parameter data (e.g., mortality or population abundance). Anatomical placement, seasonal fish activity (such as spawning), and fish size may also affect PIT tag retention [6]. PIT tags can be implanted subcutaneously into the dorsal site or the peritoneal cavity, with some researchers recommending sutures or adhesive to heal entry wounds [6, 60]. Some field studies of estuarine fishes with PIT tags incorporate mortality, survival, growth and tag retention data from other species to support tag loss assumptions [42].

Few studies have compared retention rates, even though many have found the rate of retention at this anatomical position. PIT tags that were inserted into the peritoneal method was found to increase survival when compared to injecting tags into the belly with a syringe and plunger [4] (Fig. 3).

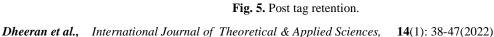


Fig. 4. Tag detector.

cavity have reportedly been found to be shed tags by mature fish at the time of spawning [58]. Because of this, retention rates may decline during the peak spawning seasons, however, little study has examined this. The size of the fish may also have an impact on tag retention [6].

Finally, controlled laboratory or aquaculture protocols account for the majority of PIT tag retention estimations. However, they might not accurately simulate environmental elements that affect retention, such as increased swimming during floods, the effects of heat on the healing of the tag insertion site, and the rigours of spawning. Additionally, earlier PIT tag studies have shown strong retention and identification rates, which are the two fundamental tenets of the majority of acquisition models [56]. It is not usual for field studies on estuarine fishes to provide no information on the target species' development, survival, or tag retention [30].





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C. Factors Needed to be Considered While PIT Tagging to Minimize Stress

A). Fish Health. When it comes to evaluating stress levels following tagging, the health of the fish being tagged is crucial. Fish that are infected with a bacterial, viral, or fungal disease should not be used for tagging, and the survival rate will be low due to the stress involved with the tagging procedure. PIT-tagging activities should be postponed until the illness outbreak is under control, or healthy fish should be used for tagging [57].

B). Temperature. Increased temperatures reduce tagged fish to cope with stress. It is necessary to maintain optimal tagging temperatures for both cold and warm water fish. Tagging coldwater fish below five degrees Celsius has little effect on the fish. Coldwater fish are easily stressed when the temperature rises above 15 degrees Celsius. During tagging fish in temperatures above 15°C, precautions should be taken to avoid mortality. Monitor the fish in holding tanks with proper aeration and in anaesthetic baths with proper concentration, and stop operations immediately if fish begin to show indications of stress. It is not suggested to tag coldwater fish in temperatures over 17°C, and handling fish in temperatures above 20°C must be avoided. To avoid working with increased temperatures, tagging should be performed early in the morning when temperatures will be lowest or wait for a suitable temperature if you're tagging coldwater species [57].

C). Oxygen. At very low oxygen levels, fish stress increases. It is critical to provide continual aeration to recovery tanks and anaesthetic baths. Because warm water holds less oxygen, oxygen becomes increasingly vital as the water temperature rises. There are two methods for obtaining oxygen. One method is to use air or oxygen to bubble through the water. An air pump and air stones can be used to provide air. Oxygen cylinders can be used to provide oxygen. Running fresh water through your recovery tank is another approach to provide oxygen. In the case of Coldwater species, it helps to keep the temperature in the recovery tank similar to that of the stream [57].

D). Fish Handling. Fish stress levels during tagging can be influenced by how you handle them. When scoop-netting fish, try to avoid chasing them around. Avoid catching too many fish at the same time in the scoop net for tagging. The experimental set-up should be ready so you can catch fish for sedating in an anaesthetic bath immediately. If you need to transport fish over small distances, use a sanctuary net. As the water temperature rises, stress management becomes more consideration. Avoid handling fish twice. Stress builds up over time, therefore if you disturb the same fish several times at the same time, the stress level of the fish will increase [57].

E). Aggregation. Increased density can also lead to increased stress levels in the tagged fish, especially if you rearing too many tagged fish in the rearing tank.

Decrease fish density if too much dense in the rearing tank. If aggregation of fish occurs in the rearing system, avoid increased density of tagged samples. Observe crowded fish continuously for signs of infection. During several situations under hatchery conditions, where excess numbers of fish have suffocated one another in such instances. Mass mortality will occur during such situations. It is very much necessary to continuously monitor tagged fish that has been kept under increased density during rearing conditions [57].

Anesthesia. Anaesthetics are required in **F**). aquaculture to decrease handling stress as well as mortality. Traditional anaesthetics such as tricaine methanesulphonate (MS-222), 2-phenoxyethanol and quinaldine, are expensive and are poisonous to fish. The ideal anaesthetic should have a short induction time (1-5 minutes) and a short recovery time (less than 5 minutes), as well as being inexpensive, simple to apply, easily soluble, and should not leave any residues in fish, humans, or the surrounding environment [15, 68]. The FDA has approved MS-222, also called tricaine methane sulfonate (MS-222), as an effective anaesthetic for fish as well as other cold-blooded animals. The most efficient chemical for anaesthetizing salmonids is neutralised MS-222 (pH 7), but at the time of increased concentration or the fish is sedated in the anaesthetic bath for a longer period, it can cause injury or death [69]. MS-222 can cause a state of hypoxia by decreasing the opercular movement to alter the flow of water across the gills, so it will reduce the exchange rate of oxygen between blood and the water. If fishes are kept under anesthetization for a longer period, they may experience hypoxic conditions, which can result in permanent brain damage or death [57].

i. Concentration. To anaesthetize salmonids, a concentration of MS-222 of around 40 mg/l is indicated [61]. The concentration of anaesthetics will vary depending on the environmental conditions, the fish species, the size of the fish, and the level of stress. As the temperature increases, the metabolism of the fish will also increase, which means the absorption of anaesthetics occurs more quickly. As a result, fish require less MS-222 when the water temperature is warmer. Coldwater species are more susceptible to MS-222 than warm water species, requiring less MS-222 to anaesthetize them. The clove oil dosages vary from 2.0 -150 mg/L. Clove oil is an important anaesthetic for common carp Cyprinus carpio at 40-120 mg/L [17]. The sufficient dosage of clove oil to transport Oncorhynchus mykiss, is as low as 2-5 mg/L, whereas the sufficient dose for surgical an aestheticis from 40-60 mg/L. The method fish react to the anaesthesia is also influenced by their level of stress. Fish under stress conditions will need a low concentration of anaesthesia to anaesthetize fish for tagging.

ii. *Stock Solution.* It is necessary to prepare an anaesthetic stock solution to make anaesthetics proper soluble to drug the anaesthetic bath. Using this procedure, the proper anaesthetic concentration can be

effectively used by the fish and the chance of overdosing on the anaesthetic bath can be reduced. The concentration of the stock solution can vary slightly depending on your preferences, but for MS-222, the concentration should be between 40 and 50 mg/l. The suggested concentration for MS-222 is roughly 40 mg/l, which needs about one millilitre of the stock solution for one litre of water in the anaesthetic bath. One thing to remember about MS-222 is that it is photosensitive and will decay if exposed to light, therefore you must store it in a light-proof container. You can use black electrician's tape to wrap a clear plastic container to prevent light penetration or use brown or black plastic containers [57].

iii. Anaesthetization. Initially, a low concentration of anaesthetic is used and anaesthetizes only a few numbers of fish. Keep an eye on their reaction to the anaesthesia to see if the dosage is correct. If necessary, you can easily apply additional anaesthesia. One to three minutes should be allowed for induction (the time taken for the fish to lose their equilibrium and stay on their sides). Significant operculum movement and slight fin movement should still be visible. As you work with the fish, keep an eye on them. If the opercular movement becomes weak or loses its balance, immediately remove the fish from the anaesthetic bath or keep the fish in freshwater. Death is imminent when the operculum stops moving. The fish will suffocate in minutes if there is no water circulation across the gills. Within five minutes of putting sedated fish in a recovery tank, the fish should start to regain balance and should maintain a normal swimming position. Reduce the concentration of anaesthetics in the anaesthetic bath if recovery time exceeds five minutes. The researcher suggests putting a suitable number of fish in the anaesthetic water at once so that tagging and collection of data can be finished within a few minutes of the fish being sedated using anaesthesia. In basic terms, while anaesthetizing the fish should not be kept for more than five minutes in the anaesthetic bath. Fish should not be permitted to be in an anaesthetic bath for a longer period under any circumstances [57].

G). Fish Size. The size of fish plays an important role in tagging depending on species and rearing strategy. Juvenile fish are difficult to tag, and marking them will make them more vulnerable to predators and reduce their swimming endurance [46]. Furthermore, some studies have discovered that compared to large fish, fewer small fish are likely to be interrogated at dams [1]. The effective tag size for tagging huge fish is between 80-150 mm. The needle will easily puncture the body wall of this size fish, making it easy to handle. Small fish are difficult to grasp with one's hand, thus the insertion point may need to be changed forwards lightly to the posterior position of the pectoral fin. Hence it will provide the tag with a bit huge space in the peritoneal cavity. When working with small fish, extreme caution must be exercised to avoid hitting internal organs or intestines while tagging. Large fish with a size greater than 200 mm is hard to handle, especially with smaller hands. Huge fish are notoriously difficult to pierce with a tag injector. When the needle's point collides with a scale while tagging, the scale attaches to the injector, preventing it from penetrating the body. Take the needle out from the fish body in this circumstance, clear the scale from the injector tip, and then pierce the tag into the fish where the scale was removed from the body wall.

H). Tag Size. PIT tag's effectiveness was limited in the small-bodied fish because of their size [5]. The popular 12-mm-long tag has been replaced by 9-8mm small tags, with 9mm tags being commercially accessible in 2004 and 8mm small tags in 2014. These smaller tags are allowed for tagging smaller fish but their read ranges are shorter, which could be difficult to detect tagged fish using remote antennas [5]. On the other hand, smaller tags have been used in field experiments for habitat usage with success [23]. Tagging juvenile fishes will help us to learn more about the ecology of a variety of fish species and their size ranges, including nongame and game species as well as concern for species conservation. Furthermore, the ability to tag small age groups of fishes would allow us to understand their site fidelity, habitat use and migrations [59].

I). Stressor. Procedures like tagging, handling, and capturing fish will generate physiological and behavioural responses in fish, and the tagged fish requires time and the proper maintenance for quick recovery. Some individuals may not be able to handle the stress of tagging, and others may find it difficult to wear the tag [67].

J). Fish Recovery and Release. Fish should be given at least a half-hour to recover in a cool, dark tank before being released back into the water source. When the tagged fish are released, they must have recovered completely from anaesthesia to escape from predators. If tagged fish are placed back into the pond ecosystem or other system, before they should have recovered from the anaesthesia in a proper hatchery condition, they may be preyed upon by their peers. During summer, some of the researchers will catch fish in the afternoon section, keep them overnight, tag them the next morning, and then release them back in the evening. This gives the tagged individuals a chance to escape from the stress of being captured or tagged before moving on to the stress phase [57].

D. Importance of PIT Tagging

Experiments in fisheries to determine the tool's usefulness in tracking fish movement and their behaviours led to the development of PIT tags in wild species in biology. PIT tags have been employed in research on reptiles [48], invertebrates [55], mammals [11] and amphibians [55]. A biological field study was applied to zoos and private collections [71] as well as the live-animal trade [29]. CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) also utilises PIT tags to verify that an

animal is captive-bred rather than wild-caught and to trace illegal animal harvest in international trade [71].

E. Effect of PIT Tagging on Movement, Behaviour, Growth and Survival

PIT-tagged animals recaptured after earlier captures may provide useful information on growth rate and position changes. Recapturing previously tagged, particular individuals in an ecosystem is also a most important technique for examining an individual's mortality and lifespan in the wild. Individually tagged species can aid in the examining of age structure and sex ratios at the species level, as well as the data needed to build demographic characteristics of populations and other life tables of the species. By using timesequenced observations of activity patterns at specific locations, researchers can investigate social networks and the behavioural interactions across species within the population [12]. PIT tags are widely used by researchers to estimate fish movement, growth, survival, mortality and exploitation studies of the tagged fish species, including the Gulf sturgeon Acipenser oxyrinchusdesotoi [12], Pacific lamprey Lampetra tridentate [49], Atlantic salmon Salmo salar [60] and brown trout Salmo trutta [18]. PIT tags are frequently employed due to their low cost, increased longevity, capacity to identify individual fish, simplicity of use, and minimal effect on survival and growth [72]. Movement behaviour, which is an important part of ecology, influences individuals' spatial interactions between different species and their environmental surroundings [51]. Fish migrate to find mating, acquire resources (such as station maintenance and range), avoid excessive conspecific density, and avoid predators (such as dispersal). The ecological and evolutionary effects of individual migration include those on population demography, individual fitness and longevity, nutrient flow throughout an ecosystem, metapopulation dynamics, species abundance and distribution. Gene flow, speciation, and adaptation are impacted by movements like dispersal [33]. Individual, group movement and population studies are unique in that they can look at processes at several levels [21]. Many systems still don't understand movement, despite its importance in analysing evolutionary patterns and ecological processes. This is particularly true in the early years when a large number of taxa comprise the dispersing class [10]. Fish migration in the river ecosystem [72] as well as through fish ways has been tracked using PIT tags and stationary tracking systems [72]. They have been employed in the lab to test the effectiveness of various clupeid fish way designs and to evaluate the swimming capability of several species [34], including Centrarchidae and Cyprinidae [62]. Experiments with smaller tags found that 8mm tags on 640-mg (wet mass) *Oreochromis niloticus* and 40–49mmFL *Oncorhynchus tshawytscha* had slightly decreased growth rates for the first 4-7 days, but increased survival (93.3%-100%) and the retention of tag (95% at 28d; 96.6% at 35 d). As a result, data from individuals who have been harmed by tagging can lead to inaccurate conclusions about wild fish development, survival rates, and behaviour [66].

F. Post Tagging Mortality

The degree to which fish are affected by tagging must be considered in trials using PIT tags because the act of tagging might be considered a physical stressor, a stimulus influencing hormone output, and a change in animal performance [20]. This is crucial in studies with smaller fish because PIT tagging has been associated with increased mortality rates in those species [25]. Using all available time points at which mortality was recorded, a random-effects logistic regression model was fitted to the cumulative mortality data [27]. Longterm risks could include tissue infections around the tagging site, which could be fatal [44]. The long-term health of the tagged animal may be directly affected by changes in growth, behaviour, immune system, and reproductive performance that are the result of tertiary stress reactions [70]. Survival and tag retention may be affected by how PIT tags are implanted in individual fish. Due to excessive needle entry into the body cavity, which results in haemorrhaging, researchers noticed significant early mortality in fish have given syringe injections [4]. On the other hand [5] employed a false injection (fish injected with a needle but not injected with a tag) and discovered that survival rates for Oregon Chub Oregonichthys crameri ranged from 93-100%. Last but not least, suturing tagging wounds improved tag retention, although survival was either the same or decreased compared to those who weren't sutured [7].

One of the components of tagging strategies that has received little attention is the tagger's or surgeon's experience with increased survival or tag retention [24]. Much research is required to determine the impact of tagging technology on wound closure in juvenile fishes because results have varied among species, sizes and studies. The impact of PIT tagging on salmonid survival has been studied in both the lab and the field [60]. *Labeo rohita* [45], *Perca fluviatilis* [7] and *Oreochromis niloticus* are just a few of the cultivated species that have been the subject of a laboratory study on the impact of tagging on non-salmonid fish survival [6, 45].



Fig. 6. Post-tagged mortality due to infection.

CONCLUSION

The PIT tag is a reputable, secure, and safe way to identify commercially significant species. It is currently the best tag for mass tagging individual fish, despite being relatively expensive. The PIT tag can be used once more after disinfection. Through effective management practises, it is possible to increase tagged fish survival rates in hatcheries, leading to increased tag recovery and fewer tag losses. Therefore, it appears that the PIT tag is currently the most palatable technique for tagging migratory species, especially for endangered species with proper tag size and size class juveniles.

FUTURE SCOPE OF THE STUDY

Knowing the entire tagging protocols can lead to finding a better position for tagging commercially important fishes to achieve increased seed production through selection.

Acknowledgement. We thank the support rendered by the concerned departments of Aquaculture, Kerala University of Fisheries and Ocean Studies, Panangad, Kochi 682506, Kerala, India during the investigation.

Conflict of Interest. All authors declare that they have no conflict of interest.

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