

## New Trends in Control of Parasitic Infections in Marine Fish

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**Abstract:** Parasitic diseases of fish are very important in aquaculture as it was the basis for all outbreaks of fish. It is very important to accurately diagnose and control of the parasitic diseases of fish to avoid great economical losses in aquaculture. So it is very important to discover new techniques for diagnosis and control of fish parasitic infections. Traditional chemotherapeutic control for fish parasitic infection must be avoided for its great harmfulness like accumulative residual in fish flesh and contamination of the environment, for these reasons new techniques must be developed for control of parasitic diseases of fish. One of the most essential methods are vaccination against parasites of fish, administration of dietary probiotics and immunostimulants like spirulina and ascorbic acid for enhancing the innate and acquired immune response and weight gain in fish. Also, using of medicinal plants as anti-parasitic and immunostimulating materials. Biological control of fish parasites using cleaner fish and shrimp for removal of fish external parasites, cells and mucus from the external body surface. Lastly, the most important method for control of fish diseases is what is called “biosecurity” for prevention or reduction of the entrance of pathogenic organisms into the aquatic culture by including; sanitation, disinfection and quarantine.

[Hussien A.M. Osman and Mona S. Zaki. **New Trends in Control of Parasitic Infections in Marine Fish**. *Stem Cell* 2018;9(1):37-53]. ISSN: 1945-4570 (print); ISSN: 1945-4732 (online). <http://www.sciencepub.net/stem>. 7. doi:[10.7537/marsscj090118.07](https://doi.org/10.7537/marsscj090118.07).

**Key words:** New Trends, treatment, control, chemotherapeutic, probiotics, immunostimulants, Biological control, biosecurity.

### 1. Introduction:

Parasitic diseases form about 80% of fish diseases. This might be because of the long time of warm climate and wealth of characteristic sustenance and also the accessibility of the main intermediate hosts like cyclops, mollusca and parasites. The parasite is the premise of all outbreaks of fish that took after and showed by optional disease with microorganisms or organisms prompting serious monetary misfortunes spoke to as high morbidity and mortality (Eissa,2002).

Parasitic diseases can affect on aquaculture systems in a number of methods which will determine their economic cost (Sommerville, 1998; Lagrue and Poulaine, 2015). Mortality has evident costs determined by the size and age of the fish, but, more often, parasite infection causes morbidity and loss of appetite with a resultant waste of food, reduced food conversion efficiency and specific growth rate, which over a grow-out period in a population of fish, may account for a noteworthy proportion of the profits (Trujillo-Gonzalez, *et al.*, 2015).

In some cases, parasite infections might be zoonotic, or reduce the market value sector esteem inferable from their expansive size or unaesthetic appearance, and this has been known not dismissal of whole loads of fish even where there was no detectable effect on the welfare of the fish; other parasitic diseases may influence brood stock quality (Santoro *et al.*, 2013; Timi and Mackenzie, 2015).

A common mistake of fish culturists is misdiagnosing infection problems and treating their diseased fish with the wrong drug or chemical. At the point when the substance doesn't work, they will attempt another, then another. Selecting the wrong treatment in light of misdiagnosis is a waste of time and money and might be more hindering to the fish than no treatment by any stretch of the imagination. The greater part of fish parasites must be distinguished by the utilization of a microscope. In the event that the microscope, or the individual utilizing it has no past experience, the determination is troublesome and faulty (Noga, 2010).

Regarding control of parasitic diseases there are new approaches dealing with such point in addition, to the known traditional methods for control of fish diseases, such as immunestimulants, probiotics, prebiotics, vaccines, herbal medicine and biological control. Besides, biosecurity which is the more advanced scheme developed recently in fish farms (Sommerville, 2012).

Thus, the present state of the art was aimed to throw light Hutson *et al.*, 2012; Muktar *et al.*, 2016) on the new trends of diagnosis and control of marine fish parasitic infections like molecular techniques for the detection and identification of parasitic fish pathogens. Also transmission and scan electron microscope and displaying the new trends in control parasitic fish infections using immunoprophylactic control like vaccines, immunostimulants, probiotics as

well as physical, mechanical and biological control in addition to herbal medicine.

#### **A. Traditional or chemotherapeutic control of fish parasitic diseases:**

A wide range of chemicals was used in the aquaculture industry.

-Before chemotherapeutic treatment is connected, it is important to make exact diagnosis of the problem (Woo,2012).

-On account of a parasitic problem, it may not be conceivable to make a complete distinguishing proof of the parasite. In any case, in any event the group of the parasite must be known (Noga, 2012).

-A decreased volume of water. This offers the joined advantages of having the capacity to rapidly bring the water level up so as to weaken the treatment in the occasion of an antagonistic response and diminish the expense of chemicals utilized (Eissa, 2002).

-It is additionally fundamental to watch the fish during the treatment to be prepared to make any important emergency action (Noga, 2010).

#### **A.1. Disadvantages of chemotherapeutic treatment of fish parasitic diseases:**

1- A few chemicals have cancer-causing activity like malachite green on fish and human, low withdrawal time and remaining viability on fish substance. It might accumulate hurtful residues in the fish flesh living and the environment (Zhan and Braunbeck, 1995).

2- Some chemicals must be used at low water temperature e. g formalin. It reduces the dissolved oxygen level in water (more high temperature low oxygen level in water). Vigorous aeration should always be provided. Also, it is not used in food fish, used only in ornamental fish (Fajer-Avila *et al.*, 2003).

3- Other chemicals must be applied to water pond with low organic matter like potassium permanganate due to the organic matter oxidizes making it usefulness. It is not recommended as it can burn gills of fish and aeration should be available when potassium permanganate is used because it is an algaecide and can cause oxygen removal (Eissa, 2002).

4-When using any treatment for fish a bioassay (a test to determine safe concentration) should be carried out on a few fish before large numbers of fish are *exposed* (Pironet and Hones, 2000). Generally we must always remember that chemotherapeutic control of fish pathogens is the last trial for control, as most chemicals used for treatment affect the parasites as well as the fish.

#### **B. New trends in control of parasitic fish Infections:**

##### **1. Physical treatment of fish parasites:**

There are some atypical methods for treatment some types of fish parasites such as: This type of control is used for breaking down the life cycle of parasitic infection in different stages. It is used for example in treatment “white spot disease”.

##### **1.1. Flushing method:**

It is utilized for the most part as a part of incubation facilities or tanks. The fish are held in containers with punctured base and sides for three weeks and kept up in running water. This procedure may prompt the expulsion of all stages of the parasite. As the parasites leave the diseased fish are flushed out with water, this will cut their life cycles (Noga, 1996).

##### **1.2. Transferring method:**

It is used for ornamental fishes as the infected ones are transferred to parasite free aquaria every day or 12 hours. The tomonts fall down into the first container developing into theronts. The repeated transfer of fish to parasite free aquaria is continued for one week (Eissa, 2002).

##### **1.3. Heat method:**

This technique depends fundamentally on raising water temperature up to 30 to 32°C. It must be raised gradually and not more than 1°C/hour and kept up for 5 to 7 days then brought down gradually once more. It must be utilized just for tropical fishes particularly in some ornamental fishes. By putting the infected fishes in a tank or aquarium without aquatic plants with high temperature, the parasites encyst separating by straightforward direct divisions or even leave the bucket from absence of oxygen. Thus, the high temperature is not the explanation behind murdering the parasites but rather it acts in a roundabout way by the absence of water oxygen, which can come about because of it. Likewise, the higher temperature the faster the parasite will develop to develop size and leave the fish to the unfavorable encompassing water (Eissa, 2002; Noga, 2010; Brazenor and Hutson, 2015).

#### **2. Mechanical treatment of fish parasitic infections:**

These methods are used for treatment and control of crustacean parasitic diseases of fish for example:

##### **2.1. Argulosis:**

They can be removed manually with a blunt forceps or soft brush, when few fish of high value (spawners and ornamental fishes) and when only a limited number of fish are involved.

Embeddings sticks or wooden boards (1x0.5m) submerged in the lake (having a profundity 0.75m) as female arguli will leave fish and store their eggs on the boards that go about as egg - trap sticks. They are expelled, inspected, cleaned of following eggs and supplanted each day or twice week after week. This will devastate incredible quantities of Argulus eggs particularly in little lakes (Eissa, 2002).

## 2.2. Lerneosis:

Transformed female *Lernaea* can be separated with forceps and expelling it off with a couple of scissors. The parasite is killed and advancement of its egg is avoided. The egg sacs must not be come back to the water. This methodology should be trailed by neighborhood treatment with cotton swabs pervaded with reasonable antifungal. This strategy is generally used to treat *Lernaea* infestation of aquarium fishes and profitable ones, for example, spawners (Eissa, 2002).

### Immuno Prophylactic Control:

**2.3. Probiotics (Yeasts e.g. *Saccharomyces cerevisiae*).**

**2.4. Immunostimulants (e.g spirulina and ascorbic acid).**

**2.5. Vaccination against fish parasites.**

Immunoprophylactic control of fish infections includes pro and prebiotic medications, immunostimulation and established vaccinology, all preventive measures aimed for production compounds help the bactericidal action of phagocytes (Ellis, 1999).

The old meaning of the term probiotics is: "a live microbial food supplement which helpfully influences the host by enhancing the intestinal balance" (Fuller, 1989), the importance focused on "live" and "intestinal". Practically speaking, nonetheless, the term has likewise been connected to non-viable microbes, not inexorably limited to the intestinal parity. An immunostimulant has been characterized by Bricknell and Dalmo (2005) as: "an actually happening intensify that tweaks or improve the safe immune system by increasing the host's resistance against diseases that it is created by pathogens" (Kesarcodi-Watson *et al.*, 2008).

### 3. Probiotics for fish:

Probiotics are live, microbial, dietary supplements which stay alive as they enter the gastrointestinal tract and enhance the host's wellbeing (growth and immune response) (Gatesoupe, 1999). Their helpful impacts include: grip to the intestinal tract, creation of inhibitory compounds, rivalry for chemicals and immuno-stimulation (McCraken and Gaskins, 1999).

The utilization of probiotics is settled in domestic animal farming and has as of late likewise been utilized as a part of aquaculture (Sharifuzzaman and Austin, 2009).

Commercially accessible probiotics, for instance, lactic acid bacteria, *Bacillus* sp. in addition, yeasts, are regular fixings in human wellbeing nourishment and in domestic farming animals. Some of these have been tried in aquaculture with by and large great results (Ma *et al.*, 2009).

A few commercial probiotic preparations have

also been developed specially for use in aquaculture like MacroGard (B-glucans) and Ergosan (polysaccharide) from seaweed (Bagni *et al.*, 2005).

A quest for probiotic microorganisms in aquaculture incorporate the screening of the bacterial flora connected with the fish under study and/or its surroundings. The antimicrobial activity against fish pathogens is then tried, the pathogenicity in the host fish is checked lastly the consequences for development and survival of fries or tested fish are inspected (Kesarcodi-Watson *et al.*, 2008). A few experiments of this nature have been portrayed in the writing where the deciding item has demonstrated accommodating for the fish (Sharifuzzaman and Austin, 2009). There is also some types of probiotic microbes isolated from the host or its surroundings are more valuable than commercially accessible products (Lauzon *et al.*, 2009).

Live probiotic bacteria are accepted to influence the intestinal flora, their adversarial action and rivalry for supplements and space diminishing the quantity of pathogenic microbes. They are also thought to enhance ravenousness and advance development. There are case of probiotic treatment upgrading cellular and humoral resistant parameters (Irianto and Austin, 2002b). Probiotics may also fortify characteristic protection other cell receptors or humeral figures however for this to happen the probiotic microorganisms would need to pass the mucosal and epithelial barreiers of the wholesome trench and deliver the normal defense mechanisms of the fish (Planas *et al.*, 2006).

#### 3.1. Yeasts as probiotic for fish:

Yeasts are most critical microorganisms, which spread with animals, air and water streams, and can develop in different situations where natural substrates are accessible. Their presence has been noted in fish guts for quite a while in wild, and additionally cultivated animals, however this common event has been by and large considered as coincidental. Modern yeast is ordinarily utilized as a part of aquaculture, either alive to nourish live micro-organisms, or in the wake of preparing, as a food fixing (Stones and Mills, 2004).

Yeasts can stimulate the immune response in fish.  $\beta$ -glucans are likely the most essential mixes in such manner, yet some other cell-wall segments or dissolvable variables may also assume a part. Both cell and humeral responses have been evoked by dietary yeast, contingent upon the test conditions. Different advantages might be normal for the host, particularly the intestinal colonization of early nourishing fry with yeast, which may have some impact on improvement, e.g. by quickening the development of the digestive system. In older fish, dietary yeast may animate digestion system and development Gatesoupe (2007).

The impacts of two yeasts on European sea bass fries feed compound diets. *Debaryomyces hansenii* HF1 enhanced survival, and vertebral compliance of the fries, perhaps because of the watched speeding up of the development of the digestive system. These impacts were not saw with *S. cerevisiae* X2180, and the larvae developed better when they were encouraged a control diet, likely because of a lacking method for presentation of the yeasts after food preparing, which crumbled the physical properties of the pellets (Tovar *et al.*, 2002).

Dietary spermine additionally actuated intestinal development in European sea bass larvae (Péres *et al.*, 1997). Therefore, polyamine secretion was a conceivable mediator for the impact of the yeast on rainbow trout fries (Waché *et al.*, 2006). *D. hansenii* HF1 delivered a great deal more spermidine than *S. cerevisiae* X2180, and that may likewise represent the impacts present in European sea bass (Tovar *et al.*, 2002).

Yeasts are especially intriguing in light of the fact that they give  $\beta$ -glucans and nucleotides that stimulate the immune system of fish (Sahoo and Mukherjee, 2001). The yeast *Debaryomyces hansenii* was disconnected from the gut of rainbow trout raised in new water. Not just is it a polyamine-creating yeast fit for holding fast to fish intestinal mucous, yet it additionally constitutes a standout amongst the most essential types of yeast (Tovar *et al.*, 2002). An expanded of the survival and digestive enzyme activity in *Dicentrarchus labrax* larvae fed diet containing *D. hansenii* (Tovar *et al.*, 2004).

The utilization of commercial baker's yeast, *Saccharomyces cerevisiae* as a development and safety promoter for Nile tilapia, *Oreochromis niloticus* (L.). The development advancing impacts of baker's yeast were seen with fish and the ideal development, encourage use, and protein turn-over were gotten with 1.0–5.0 g yeast/kg diet. Notwithstanding, the most minimal fish mortality and bacterial include were acquired fish sustained 5.0 g yeast/kg. These outcomes show that baker's yeast supplement is promising as a different option for anti-microbials for disease counteractive action in tilapia aquaculture (Abdel-Tawab *et al.*, 2008).

Baker's yeast, *Saccharomyces cerevisiae*, is utilized for the baker's industry that contains different immunostimulating compounds, for example,  $\beta$ -glucans, nucleic acids and in addition, mannan oligosaccharides, and it has the capacity to advance immune response (Ortuño *et al.*, 2002) and development (Li and Gatlin, 2004) of different fish species. Be that as it may, the organization of yeast has been perceived to have essential impacts as immunostimulant (Osman *et al.*, 2010).

#### 4. Immunostimulants for fish:

The term immunostimulant covers an extensive variety of substances that improve the immune system example acknowledgment proteins/receptors and TLRs and upgrade general defence to different diseases. Immunostimulants might be bacterial or plant substances and also some synthetic compounds (Robertsen, 1999).

Immunostimulants a progression of auxiliary particles from different sources e.g. microbes, yeast, fungi and lichens alone or together with synthetic products (e.g. oligonucleotides) is right now utilized as immunostimulants. An expansive group of compound on this item retire is the beta-glucans (Skov *et al.*, 2012).

The immunostimulants have pathogen related molecular patterns (PAMPS) which can animate distinctive pathogen acknowledgment receptors (PRRs) in host fishes and in this manner activate immune responses (Chettri *et al.*, 2011). These involve among others toll-like receptors which accordingly can start course responses which may restrain or decrease diseases infections. Results from controlled research facility (Jaafar *et al.*, 2011) or field thinks about (Xueqin *et al.*, 2012) have shown that invulnerable components are enacted by these food added substances. Lamentably the impact of these food added substances on parasite infection levels, for example, intensities of the skin parasitizing ciliate *I. multifiliis*, is constrained. Correspondingly, immunostimulant encouraging does not bring about an unmistakable diminishment of *L. salmonae* disease in salmonids yet simply prompts a more favorable course of infection (Guselle *et al.*, 2010).

One of the most used substances in immunostimulation experiments in fish are various forms of  $\beta$ -glucans from different sources, normally introduced in the feed but also by intraperitoneal injection or as vaccine adjuvant (Dalmo and Bøgvold, 2008).

The increase of vitamins, carotenoids and herbal remedies for the fish feed have been tried in aquaculture. The impacts have been useful, for instance, decreasing stress reaction, increasing the activity of inalienable parameters and enhancing disease resistance and protection (Yin *et al.*, 2009).

##### 4.1. Spirulina (*S. platensis*) as immunostimulant

Spirulina *S. platensis* was as characterized a blue green micro algae that has been utilized since old times as a wellspring of nourishment as a result of its high healthful worth (Dillon *et al.*, 1995). Spirulina (*S. platensis*) is a blue-green filamentous algae that develops in carbonate-rich lakes in Torrid Zones. Spirulina is a crisp water microalga of the class Cyanophyceae, is a decent wellspring of protein and vitality. It can somewhat supplant fish feast protein in

fish nourishes and can be administrated to deliver key amino acids, vitamins, regular  $\beta$ -carotene, and antibacterial substances of good quality and quantity (**Harel et al., 2002**).

The cyanobacterium *S. platensis* is by and large generally concentrated on, for nourishing reasons as well as for its restorative properties; along these lines, a few studies have demonstrated that Spirulina or its concentrates could avert or repress tumor in animals, and the works have shown that this species has immuno-advancing impacts (**Subhashini et al., 2004**).

Spirulina is a freshwater microalga. A few botanists have characterized Spirulina as a cyanobacterium. *S. platensis* is utilized broadly as a part of dietary creation, because of its potential antiviral, cancer prevention agent, hepatopreserver, antiallergenic and immunomodulator activities (**Khan et al., 2005**).

The immunostimulatory impacts of the dietary spirulina (*S. Platensis*) was tested on tilapia, (*Oreochromis niloticus*); After the fish were nourished with Spirulina and the parameters of non-specific immune response, including serum bacterial action, phagocytosis and lysozyme action were performed following 4 weeks. The survival was also dictated by bacterial test and observing particular neutralizer levels (Micro-agglutination), the outcomes demonstrated that Spirulina improved reactions of bactericidal, phagocytic action and lysozyme action (**Khalil et al., 2007**).

Cyanobacteria (blue green growth) are photosynthetic prokaryotes utilized as nourishment added substance. They have additionally been perceived as a rich source of vitamins and proteins and all things considered are found in wellbeing sustenance stores world wide (**Singh et al., 2005**).

#### **4.2. Ascorbic acid (vitamin C) as immunostimulant:**

It was clear that 19.5 mg ascorbic acid (AA)  $\text{kg}^{-1}$  was adequate to prevent insufficiency signs and enhance survival of golden shiner in aquaria, while alternative complement activity and imperviousness to warmth stress were expanded by increasing dietary vitamin C levels. Subsequently, the dietary prerequisite for AA differs with metabolic capacity in this species (**Chen et al., 2003**).

Vitamin C (or AA) and vitamin E function as organic cancer prevention agents to ensure cell macromolecules and other cell reinforcement particles from uncontrolled oxidation by free radicals during typical digestion system or under the states of oxidative challenge, for example, disease and anxiety. Because of their potential for communication, dietary necessities for vitamins C and E are regularly viewed as together in Atlantic salmon (**Hamre et al., 1997**) and rainbow trout (**Furones et al., 1992**). Fish fed

high concentration of dietary AA accumulated high tissue concentration, which may give extra protection against oxidative damage from environmental stress and disease infections (**Lim et al., 2001**).

Fish had diminished rate and seriousness of vitamin-E-inadequacy signs with expanding dietary AA level, as found in the rate of fish with darkened skin, hematological parameters, alternative complement activity, and cumulative mortality taking after introduction to warmth stress. Be that as it may, dietary AA did not forestall vitamin-E-insufficiency signs in golden shiner (**Chen et al., 2004**).

Vitamin C promote a vital part in immunity and growth of fish (**Lin and Shiau, 2005**). Fishes can't synthesize ascorbic acid because of the absence of L-gulonolactone oxidase that is in charge of synthesis of vitamin C (**Fracalossi et al., 2001**). Subsequently, an exogenous source of vitamin C is required in fish diet. Inadequate supply of dietary vitamin C as a rule results in various lack manifestations like spinal distortion, internal haemorrhaging, retarded growth and depressed immunity (**Gouillou-Coustans et al., 1998**).

Vitamin C has been imagined that to be a standout amongst the most imperative supplements identified with fish safety (**Ai et al., 2004**). Various past written works demonstrated the valuable impacts of high vitamin C consumption on immunological parameters, for example, lysozyme, complement activities, phagocytic activity, respiratory burst and characteristic cytotoxic action (**Anbarasu and Chandran, 2001**), upgraded resistance to stress and diseases (**Montero et al., 1999**).

Vitamine C has additionally been appeared to be a critical supplement relating with fish immunity. The valuable impacts of vitamin C on different immunological parameters have been accounted for. Dietary AA upgraded serum bactericidal activity, phagocytic action, counter acting agent levels, serum complement activity and lysozyme action (Lin and Shiau, 2005).

**Ai et al., (2004)** reported that the activities of lysozyme and alternative complement pathway in serum advanced with the expansion of dietary vit C. At the point when dietary ascorbic acid achieved 489.0 mg  $\text{kg}^{-1}$ , these parameters were significantly increased than those of every single other treatment. Besides, **Ren et al., (2005)** reported that vitamin C is required by Japanese eel and could upgrade serum bactericidal activity when nourished an eating routine containing crystalline vitamin C. Be that as it may, no data is accessible on the impact of AA subsidiaries as a vitamin C source. What's more, **Ren et al. (2007)** reported that fish nourished eating regimens containing 762 mg AA/kg demonstrated essentially higher lysozyme action of mucous and serum, and

bactericidal activity of mucous than the fish sustained the eating regimen containing 32 mg AA/kg. Survival reaction of the exploratory fish fed on supra dietary levels of vitamins was fundamentally superior to that of the control set (Azad *et al.*, 2007).

##### 5. Vaccination against parasites of fish:

There is extensive variety of parasites in both wild and cultured fish stocks. Although parasitic diseases, for example, amoebic gill infection, white spot disease, whirling disease, proliferative kidney disease (PKD) and Salmon lice infestation make several problems in fish cultivating (Woo *et al.*, 2002), no parasite vaccines are commercially accessible (Somerset *et al.*, 2005). When all is said in done, fish have both humoral and cell-intervened immune response against numerous parasites and there are numerous reports on immunity/expanded resistance among fish surviving common parasitic infection (Ellis, 1999). Cultivation of parasites for potential killed or live vaccin is much more costly than virus cultivation (Somerset *et al.*, 2005), as a host populace instead of cell cultures are normally required. Notwithstanding the high costs, the utilization of normal hosts for development of parasite would make significant problem regarding safety documentation. Along these lines, recognizable proof and production of defensive antigens is presumably the most attainable procedure towards commercial parasite vaccines, at least for low cost vaccines (Plant and LaPatra, 2011; Muktar *et al.*, 2016).

High efficacies of anti-viral and anti-bacterial vaccines have also been demonstrated and efforts should therefore be made in order to develop antiparasitic vaccines (Lorenzen *et al.*, 2000).

Because of the restricted size of numerous protozoan parasites the cell and humoral immunological arm in fish should have the essential capacity to create defensive safety against in any event some of these pathogens. Experimental vaccines against *C. salmositica* (Woo, 2012), *L. salmonae* (Rodríguez-Tovar *et al.* 2006) and *I. multifiliis* (the freshwater proportional to *C. irritans*) (Alishahi and Buchmann, 2006) have been observed to be in part viable which is promising for further accomplishments. *Cryptocaryon irritans* is known not antibody production in affected fish which is an important premise for further vaccine improvement (Misumi *et al.*, 2011).

Experimental studies have also demonstrated significant activation of cellular and humoral immune factors in fish exposed to several other protozoans such as *A. ocellatum* (Noga, 2012) and *I. necator* (Chettri *et al.*, 2012).

Subsequently, it is likely that creation of antibodies against *A. ocellatum*, *S. salmonicida*, *I.*

*necator* and *L. salmonae* might be reasonable objectives for reestablished research endeavors. Nonetheless, it can't be rejected that the parasites themselves or parasite determined molecules redirect the reaction of the host towards a less viable invulnerable pathway and this plausibility should be considered when planning vaccines and their adjuvants. The late advancement of reagents and instruments for researching immune systems in fish can make a premise for commercial vaccines empowering immune response in fish which can give protection (Jørgensen and Buchmann, 2011).

The vaccine viability in fish is by and large settled by challenge experiments. In mammals specific antibody response is a decent marker of antibody adequacy however in fish this is questionable. Some fish species will deliver protective antibody response to vaccination (Pakingking *et al.*, 2009).

The creation of commercial anti-parasite vaccine is essentially postponed because of presentation of complex structure and life cycle of the parasites and the challenges in cultivating the parasite in adequate amount for vaccine production. Distinguishing proof of the parasite antigen that will induce defensive safety is also required. This is essential while considering recombinant protein creation, which is an appealing methodology in anti-parasite vaccine production (Chettri *et al.*, 2009).

The development of acquired immunity to various parasites for fish is now known to exist. Therefore, vaccination should be developed urgently to protect fish against parasitic diseases. (Gillund *et al.*, 2008 and Pakingking *et al.*, 2009).

##### 5.1. Vaccination against *Cryptobia salmositica*

The incubation facility raised Atlantic salmon, *Salmo salar* L., were inoculated intraperitoneally (IP) with a live lessened *Cryptobia salmositica* vaccine and 4 weeks later were challenged with the parasite. Unvaccinated, tainted salmon had high parasitaemias and were anemic. Fish given a high dose (100,000 parasites/fish<sup>-1</sup>) had higher parasitaemias than fish given the lower dosage. vaccinated fish had low parasitaemias and a gentle pallor and recouped rapidly after test Ardelli and Woo (2002). Moreover, Chin and Woo (2004) reported that *Cryptobia salmositica* is viewed as a deadly pathogen of salmon kept under semi-characteristic and concentrated environment. The live *C. salmositica* vaccine incited a trademark parasitemia profile and humeral reaction in Atlantic salmon. However, cryptobia-vulnerable *Salvelinus fontinalis* inoculated with a live *Cryptobia salmositica* vaccine were ensured against *C. Salmositica* and they were still ensured for a long time after their initial challenge (Ardelli and Woo, 2006).

The adequacy of attenuated live *Cryptobia salmositica* vaccine, the impacts of sustenance

hardship on the invulnerable reaction and its term in rainbow trout (*Oncorhynchus mykiss*) inoculated with a live *C. salmositica* vaccine or with a murdered *Aeromonas salmonicida* vaccine. The study demonstrated that the attenuated strain of *C. salmositica* did not bring about animea and disease, and the fish were avoided from clinical disease when they were challenged with virulent parasites (**Fard and Woo, 2008**). Salmonid cryptobiosis is brought on by the haemoflagellate, *Cryptobia salmositica*. The clinical indications of the sickness in salmon (*Oncorhynchus* spp.) incorporate exophthalmia, general edema, and abdominal distension with ascites, anemia, and anorexia. A single dosage of the attenuated vaccine secures 100% of salmonids (juvenils and grown-ups) for no less than 24 months (**Woo, 2012**).

### 5.2. Vaccination against ichthyophthiriasis:

Channel catfish vaccinated with live theronts or sonicated trophonts acquired protection against Ich. The immunized fish indicated larger amounts of anti-Ich antibodies in the skin and serum than non-vaccinated fish. Parasite infection and mortality did not happen in fish vaccinated with live theronts. Fish vaccinated with sonicated trophonts had a low infection and mortality compared with non-vaccinated fish (**Xu et al., 2004**). Delivered commercial Ich vaccine was produced and exhibited that the immunization against Ich incited defensive safety and could give a one of a kind answer for keep this parasitic disease (**Xu, 2010**).

### 5.3. Vaccination against *Discocotyle sagittata* (monogenea):

When rainbow trout *Oncorhynchus mykiss* were injected intraperitoneally with 2 different *Discocotyle sagittata* extracts dissolved in PBS and subsequently exposed to infection. Immunization resulted in significantly reduced worm intensities in vaccinated fish (**Godoy, 2003**).

### 5.4. Vaccination against *Loma salmonae*:

The earlier presentation of rainbow trout *Oncorhynchus mykiss* juveniles to the low-destructiveness variation of *Loma salmonae*, spores brought about a xenoma force in the gill fibers fourteen times lower than that saw in the control fish. Lessening in quantities of xenomas proposes that utilization of the low-destructiveness variations should be further considered as a technique to protect fish (**Sanchez et al., 2001**). The powerful vaccine spore dosage of a low-destructiveness strain of *Loma salmonae* to constrain microsporidial gill disease in trout, they found that fish accepting  $10^3$  to  $10^5$  executed spores had the great protection against exploratory infection, with 85% less xenomas in gills (**Speare et al., 2007**).

## 6. Biological control of fish parasites (life against life):

Biological control: "Any activity of one animal categories that decreases the unfavorable impact of another. It is the more security method for control of fish parasites, utilized for the most part as a part of control of crustacean parasites of fish. Parasites, especially crustacean are considered as an essential restricting element in the improvement of heightened fish culture. Under overloading conditions, parasite populaces rapidly develop and may bring about heavy mortality in the the stocked fish particularly in high water temperature where their life cycles turn out to be quicker (**Kearn, 2002**).

The control of Argulus is by presentation of mosquito fish (*Gambusia*) or healthy Angelfish, into mud ponds infested with Argulus. *Gambusia* has a distinct depressant impact on the parasitic populace as they feast upon larvae, particularly in ornamental fish lakes. Presentation of freshwater shrimp (*Macrobrachium* species), feeds on larvae and going about as a control predator for Argulus (**Eissa, 2002**).

Some monogeneans joined to the surface of a fish host may also be effectively perceptible by cleaner fish which can ingest significant quantities of monogeneans. Moreover, **Kearn (2002)** watched ingures in large monogeneans on beams most likely created by littler cleaner fish (**Cowell et al., 1993**), another capsalid monogenean, *Benedenia lolo*, parasitising *Hemigymnus melapterus* was subjected to predation by the cleaner fish *Labroides dimidiatus*. Along these lines future control measures may include the utilization of cleaner fish **Grutter et al. (2002)**.

There have been a few attempts to remove parasites by utilizing topical cleaner fish. Adolescent blue head wrasse, neon goby and cleaning goby could remove ectoparasitic monogeneans, e.g., *Neobenedenia melleni*, and from saltwater, refined Florida red tilapia and the cleaners' guts were found to contain three types of monogenean parasites. A comparative technique was connected to expel *Lernaea* from infected Arowana fish (*Scleropages* sp.), a standout amongst the most important aquarium fish, by utilizing *Tilapia nilotica* fry of 3-5-cm long as cleaner fish. The outcomes demonstrated that *Tilapia nilotica* were successful cleaner fish for *Lernaea* infestation which is called "topical cleaners fish" (**Cowell et al., 1993**).

Cleaning symbioses are interspecific relationship in which one or more member "cleaner" organism remove ectoparasites as well as tissue from the collection of another "host" organism. that frequently requests the communication by receiving stereotypic stances within the sight of the clenear. Albeit reported for some physical life forms (**Dickman, 1992**), such cooperations are best archived among marine reef

living beings, especially fish clients customers being cleaned by littler fishes, most prominently indo-Pacific cleaner wrasses and Caribbean cleaning gobies (Co'te', 2000). Cleaner fish remove ectoparasites (Arnal and Co'te', 2000), and additionally harmed tissue. However, they also expel and may very much want host mucous (Grutter and Bshary, 2003). Along these lines, whether this action really controls ectoparasite loads and results in a net advantage to has been the source of some contention (Morand, 2005).

Around 40 types of shrimps and other decapod crustaceans are also called cleaners of reef-related fishes (Chapuis and Bshary, 2009). Be that as it may, most records have been founded on perceptions of "cleaners" crawling on or picking at the outside surface of fishes, with numerous records construct exclusively in light of aquarium perceptions and/or seeming just in the mainstream aquarium writing. Just several studies have provided direct proof that any apparent cleaner shrimp species successfully expels parasites from fishes (Ostlund-Nilsson *et al.*, 2005).



**Fig. 1: Showing Moray eel infested with ectoparasitic copepods ( arrows ) that has entered a cleaner shrimp cleaning station. Note the red shrimp on the moray eel 's skin (Noga, 2010).**

In the Caribbean and tropical western Atlantic, the Pederson shrimp, *Periclimenes pedersoni* and spotted shrimp, *P. yucatanicus* (Palaemoni-dae), are two of the most widely recognized shrimps that are accounted for as a cleaner (DeLoach and Humann, 1999) (Fig.7), and are regularly found in relationship with anemones (Williams and Williams, 2000). *P. pedersoni* has been appeared to eat parasitic isopods in captivity (Bunkley-Williams and Williams, 1998), making it the main Caribbean species for which expulsion of parasites from hosts has been archived. Be that as it may, the scope of ectoparasites it devours and the potential for more term effects on ectoparasites is obscure. The united coral shrimp *Stenopus hispidus* (Stenopodidae) has also been for quite some time reported as a "cleaner shrimp" and is regular all through the Caribbean and tropical western

Atlantic (McCammom *et al.*, 2010).

Wrasse (Teleostei; Labridae) were initially used as cleaner-fish as a part of Norway in 1987, where goldsinny (*Ctenolabrus rupestris* (L.) rockcook (*Centrolabrus exoletus* (L.) and female cuckoo wrasse (*Labrus mixtus* L.) effectively expelled ocean lice from salmon in experimental situation (Bjordan, 1988) (Fig.8) From that point forward, wrasse have been utilized effectively as lice control specialists on commercial salmon in Norway (Bjordan, 1992) and in Shetland, Scotland and Ireland (Treasurer, 1994). The above three types of wrasse and the ballan wrasse (*Labrus bergylta* Ascanius) and corkwing (*Crenilabrus melops* L.) are found in Irish waters. These fish happen in beach front waters in ranges with rock and algal spread (Quignard and Pras, 1986). Goldsinny, corkwing and rockcook have all been observed to be successful cleaners, bringing about the improvement of another fishery to supply salmon farms (Deady *et al.*, 1993).



**Fig. 2: Showing Blue streak cleaner wrasse eat parasites that have attached themselves to the client fish (Noga, 2010).**

Presentation of black carp (*Mylopharyngodon piseus*) to Egypt which is molluscivorous fish of the cyprinidae family that normally happens in Chinese water. black carp has been brought into Egypt 1993 by the community oriented exploration program (CRSP) and was kept under isolate in lakes at the central Lab for Aquaculture Research (CLAR), Abbasa, Sharkia Egypt for bioconversion, sustenance and encouraging propensity considers (Soliman, 1997). Black carp has more profitable part against colonization of snails in repositories and aquaculture (Slootweg *et al.*, 1994). So it assumes a preventive part against encysted metacercaria in fish and shistosomiasis in human being (Awad, 2007).

## 7. Medicinal plants (Herbal medicine):

Medicinal plants and herbs contain substances known not and old civic establishments for their recuperating and curing properties, even old Egyptians were as similarly acquainted with drug store as they



were with solution, and they were additionally acquainted with medication readiness from plants and herbs. Therapeutic plants are imperative components for customary medication, the thought that specific plants had recuperating potential was known much sooner than individual found the presence of pathogens during the most recent years traditional medicine has not been restricted to particular society (**Ijah and Oyebanji, 2003**).

There have been expanded activities into the usage of conventional medications to control bacterial and parasitic contaminations in human and animal medicine (**Ekanem et al., 2004a**) also the utilization of therapeutic plants removes for treatment of parasitic diseases in fish has been accounted for in Egypt (**Mesalhy et al., 2008**) Restorative plants utilized as a part of aquaculture for treatment fish parasite and utilized as immunostimulants upgrading the immune response of fish.

### 7.1. Medicinal plants used for treatment fish parasitic infection.

#### 7.1.1. Medicinal plants as anti-protozoal parasites of fish:

**Yao et al. (2010)** specified that the ciliate Ichthyophthirius multifiliis is a standout amongst the most pathogenic parasites of fish. They concentrated on the impacts of crude extracts, fractions, and compounds from the leaves of *Macleaya cordata* against *I. multifiliis* under in vitro conditions by bioactivity-guided isolation method. In vitro antiparasitic adequacy experiments showed that sanguinarine was 100% viable against *I. multifiliis*. The restorative plants are right now utilized additionally as a part of commercial aquaculture as development advancing substances, supplements and antimicrobial specialists for anticipation and control of fish disease. It is additionally created as an ecologically stable supplement then again to utilization of synthetic for generation of organic fish (**Galina et al., 2009**).

There is a fast growing interest in screening antiparasitic substances from plants to replace antiparasitic and disinfectant alternatives. Two such plants are garlic (*Allium sativum*) and *Artemisia vulgarism*. Garlic is one of the edible plants that had a strong interest to scientists and recognized as an important medicinal plant which has a wide spectrum of actions; not only antibacterial and antiprotozoal, but also has beneficial effects on the immune systems (**Harris et al., 2001**).

The crude and crushed garlic (*Allium sativum*) at 200 mg/l could treat Trichodiniasis in eels **Madsen et al. (2000b)**. Moreover, of crude concentrate of garlic (*Allium sativum*) has been accounted for to murder trichodinids (**Madsen et al., 2000a**).

The crude extract from two tropical therapeutic

plants, *Mucuna pruriens* (Fabaceae) and *Carica papaya* (Caricaceae), were utilized for treatment of the ciliate Ichthyophthirius multifiliis which is the most pathogenic parasites of fish kept up in imprisonment. The result was a 90% reduction in quantities of *I. multifiliis* on fish after treatment in baths of each plant remove at 200 mg L<sup>-1</sup> compared to untreated controls (**Buchmann et al., 2003**). In addition, **Ekanem et al. (2004b)** specified that medicinal plants are vital components of traditional medicine in essentially all cultures and item guarantee a less expensive hotspot for therapeutics, more noteworthy precision than chemotherapeutic agents and a suitable answer for all problems. The control of Trichodiniasis and *Aeromonas hydrophila* in ponds of supplied tilapia with Garlic (*Allium sativum*) and Sheh el-baathran as restorative plants to treat fish. The results demonstrated that rough concentrates of either garlic and Sheh El-baathran significantly eliminated of Trichodina infections (**Aboud, 2010**).

#### 7.1.2. Medicinal plants as Anthelmintic (monogeneans):

Monogenean parasites are flatworms which inhabit skin, gills and inevitably eyes of fish. Monogeneans from the genus *Dactylogyrus*, *Gyrodactylus* and *Neobenedenia* are wide spread parasites influencing a large number of cultured fish (**Deveney et al., 2001; Woo et al., 2002**).

In result, a few studies have been performed as of late to evaluate the anthelmintic action of plant extracts keeping in mind the end goal to treat monogenean infections. Plant isolates like methanolic extract of bupleurum root (*Radix bupleuri chinensis*), aqueous and methanolic extract of cinnamon (*Cinnamomum cassia*), methanolic extract of Chinese flavor shrub (*Lindera aggregata*) and methanolic and ethyl acetic acid derivation extract of brilliant larch (*Pseudolarix kaempferi*) have appeared to possess 100% in vivo viability against monogenean *Dactylogyrus intermedius* when added to water of infected goldfish *Carassius auratus* (**Wu et al., 2011; Ji et al., 2012**). The impact of several extracts of Indian sandalwood (*Santalum album*) against *D. intermedius* and *Gyrodactylus elegans* on goldfish and found that chloroform extract was the best and most secure for the fishes. They also watched that bath treatment with long term and numerous organizations could take out an extraordinary extent of monogenean diseases (**Tu et al., 2013**).

Looking for a natural antiparasitic, in vivo anthelmintic activity of petroleum ether, chloroform, ethyl acetic acid derivation, methanol, and aqueous extracts of *Angelica pubescens* roots (*Radix angelicae pubescentis*), *Brucea javanica* organic products (*Fructus bruceae*), *Spatholobus suberectus* stems (*Caulis spatholobi*), *Aesculus chinensis* Bge. Seeds

(Semen aesculi), and *Pharbitis purpurea* (L.) Voigt seeds (Semen pharbitidis) were tried against *Dactylogyrus intermedius* (Monogenea) in goldfish *Carassius auratus*. The methanolic and aqueous extracts of *S. aesculi* displayed potential results and can be misused as a favored natural antiparasitic for the control of *D. Intermedius* (Liu *et al.*, 2010).

Hutson *et al.* (2012) evaluated the impact of aqueous extract from different algae on the lifecycle of the parasite *Neobenedenia* sp. They watched that disease success on *Lates calcarifer* was lower within the sight of *Asparagopsis taxiformis* (51%) and *Ulva* sp. (54%) removes in seawater compared to the control (71%). Besides, *A. taxiformis* remove hindered embryonic development of *Neobenedenia* sp. what's more, decreased hatching success to 3% contrasted and 99% for the seawater control. Barramundi (*L. calcarifer*) was fed with two improved garlic eats less carbs and watched that when fish were tested with *Neobenedenia* sp. treated group during long-term supplementation (30 days) showed 70% decreased infection achievement compared to control and short-term supplementation (10 days) Militz *et al.* (2013a, b).

## 7.2. Medicinal plants as immunostimulants for fish:

The immune system is arranged into innate (non-specific) and adaptive (specific) immune system. The innate system is the main line of barrier against attacking pathogens and their significant parts are macrophages, monocytes, granulocytes, and humoral components, including lysozymes or complement system. An immunostimulant is a substance that improves the defence systems or immune response (both specific and nonspecific), along these lines rendering fish more resistant to disease and external aggressions (Anderson, 1992; Chakraborty and Hancz, 2011).

An increasing interest for the utilization of plant extracts as fish immunostimulants has emerged in the most recent decade (Galina *et al.*, 2009; Vaseeharan and Thaya, 2013). A few studies have observed the immunological parameters after intraperitoneal injection or orally administered plant removes on unmistakable fish species and they found that treated fish indicated increased lysozyme movement, phagocytic activity, complement activity, increased respiratory burst activity and expanded plasma protein (globulin and albumine) (Düğenci *et al.*, 2003; Wu *et al.*, 2010; Yuan *et al.*, 2007). Lysozymes assume an essential part in the protection of fish by instigating antibacterial action within the sight of a complement (Harikrishnan *et al.*, 2012b).

Phagocytosis is one of the primary middle people of innate immunity to pathogens, while respiratory burst is additionally a significant effector instrument for constraining the development of fish pathogens.

increase in plasma protein, albumine and globulin, is viewed as a strong innate response in fishes. Different studies analyzed the hematological parameters which give some insight of the fish health status and they found that erythrocytes, lymphocytes, monocytes, hemoglobin and hemocrit levels fundamentally increased in fish treated with plant extracts compared to control fish (Divyagnaneswari *et al.*, 2007).

Some immunostimulants couldn't be utilized on account of different drawbacks, for example, high cost, constrained viability upon parentally administration, etc (Düğenci *et al.*, 2003). In aquaculture, chemotherapeutic operators, for example, commercial antibiotics and disinfectants are generally utilized for disease management, in spite of the fact that this is not fitting because of cost viability, ecological risks, and the antibiotics resistance created by numerous pathogens (Kruse and Soram 1994).

All plant extracts added to fish diet increased the aggregate protein level in plasma with the exception of 0.1% ginger. The most abnormal amount of plasma proteins was seen in the group fed with 1% ginger extract containing feed (Düğenci *et al.*, 2003). volatile oils, tannins, phenolic compounds, saponins, alkaloids, polysaccharides, and polypeptides were appeared to be powerful alternative for antibiotics. The screening of plant extracts and natural products for antimicrobial activity has demonstrated that higher plants represent a potential wellspring of new hostile to infective agents (Chakraborty and Hancz, 2011).

Herbals, for example, *Cynodon dactylon*, *Piper longum*, *Phyllanthus niruri*, *Tridax procumbens*, and *Zingiber officinalis* were separated with, benzene, butanol, and petroleum ether. The herbal diets significantly enhanced the survival, development, and immune response in comparison with the control group. The herbal diet eating methodologies enhanced immune parameters, for example, phagocytic activity and albumin-globulin proportion (Sivaram *et al.*, 2004).

## 8. Biosecurity in aquaculture (FAO,2003):

Biosecurity can be defined as 'the measures and methods adopted to secure a disease free environment in all phases of aquaculture practices (i.e. hatcheries, nurseries, growout farms) for improved profitability'. Biosecurity protocols are intended to maintain the "security" of a facility (i.e., prevent entry of, or reduce overall numbers prior to entry) with respect to certain disease causing organisms (parasites, bacteria, viruses and fungi) that may not be present in a particular system.

### Important components of biosecurity are:

#### 1-Quarantine

#### 2-Sanitation

#### 3-Disinfection.

#### 8.1. Quarantine:

defined as the isolation of an organism or group of organisms to prevent the introduction or spread of infectious disease, is a standard procedure that is extremely important in aquaculture. In practical terms, quarantine is a standard set of procedures that should be observed to prevent the introduction of pathogens or diseases into a population of fish, prawn and shrimp in aquaculture (FAO,2003).

**The quarantine protocols should be strictly used and should follow as many of the following protocols as are practical:**

- Testing of a sample of shrimp, prawn and fish before bringing them onto the facility.
- All-in, all-out stocking procedures.
- Isolation or separation from other populations for a period of time (depending upon species, diseases of concern, the system).
- Feeding observation and diet adjustment.
- Sampling and proper treatment.
- Reduction or elimination of infectious pathogens.
- Disease prevention strategies.

### 8.2. Disinfectants:

With the rapid increase in aquaculture practices, the need for disinfectants has also increased. Entry and growth of pathogens must be minimized through use of disinfectants in water, on tanks and equipment and on eggs. Disinfectants used in aquaculture are aimed at all types of infectious agents (including bacteria, fungi, viruses and parasites). The disinfectant must come into direct contact with the disease causing organism to kill them by releasing proper amounts of active compounds.

Biosecurity involves following strict management protocols to prevent specific pathogens from entering a system or reducing the numbers. A good understanding of pathogen reservoirs is important.

**An effective disinfectant is chosen based on:**

**Efficacy** - Proven efficacy is of major importance against the full range of viral, bacterial and fungal disease causing organisms. Particularly in aquaculture the viruses that cause diseases are extremely persistent and difficult to destroy.

**Environmental impact** - A good disinfectant must kill pathogenic organisms within a facility but must not harm organisms in the environment when released.

**Operator safety** – Any products used must be safe for staff employing the product.

### 8.3. Sanitation

Good sanitation procedures reduce the numbers of disease causing organisms present within a given system and prevent or reduce the spread of disease causing organisms from one system to another.

Recommended Sanitation and Disinfection Protocols to prevent or reduce the pathogen load in a system include:

**Proper attention to food sources:**

1. Be careful with live foods, although live or fresh foods can be a good source of nutrients, these may also be a source of pathogens.
2. Ensure proper storage (in a cool, dry location) and usage (follow manufacturers recommendations/expiration date) of manufactured feeds to prevent loss of nutrients and build up of pathogens or toxins.

**Good overall system maintenance and cleanliness to reduce environments that will favor pathogens and parasites:**

1. Good husbandry (nutrition, water and soil quality etc).
2. Regular monitoring of excess organic matter and control strategies.
3. Backwash and treatment of filters as needed to reduce organic loading in hatcheries.
4. Washing and disinfection of air and water pumps and lines in hatcheries.
5. flush sediment out of water lines as needed and disinfect them.
6. Maintain proper sanitisation disinfection strategies.
7. Keep nets and other equipment off the floor to control contamination and keep them sterile.
8. Pull dead and moribund culture organisms as soon as possible and dispose of appropriately.
9. Avoid cross contamination of equipment or water from one system to another.
10. Use disinfectants for equipment including nets and footbaths (placed at strategic locations around the facility, e.g., at the entrance and exit of quarantine buildings, hatcheries, farms and other systems).

### Conclusions

**From the present study, it could be concluded that:**

1. Aeration, proper management and nutrition should be kept in mind when we manage a parasitic disorders.
2. Probiotics, prebiotics, immunestimulants and vaccines should be used first in the treatment.
3. Traditional control should not be ignored in control of such problems.
4. Use of herbal medicine is a must as it has no side effects in control of parasitic fish diseases.
5. Biosecurity should be used effectively in problems associated with parasites in aquaculture.

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