Emerging Role of Medicinal Plants as Immunostimulants and Bio-antimicrobials in Aquaculture

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Abstract—*Aquaculture makes a significant contribution in the production of protein-rich food for human consumption. Aquaculture practices encounter many challenges, and one of the most devastating problems is disease outbreaks caused by microbial pathogens. Medicinal plants have been known as immunostimulants for thousands of years. The application of medicinal plants as natural and innocuous compounds has potential in aquaculture as an alternative to antibiotics and immunoprophylactics. The growing interest in these plants has increased world-wide because they are easy to prepare, cheap, and have few side effects on aquatic animals and the environment. A wide range of medicinal plants such as herbs, spices, seaweeds, herbal medicines, herbal extracted compounds, traditional Chinese medicines, and commercial plant-derived products has been studied in various aquatic animals. Medicinal plants as immunostimulants can be used not only against disease but also as growth promoters, stress resistance boosters and preventatives of infections. Plants are rich in a wide variety of secondary metabolites of phytochemical constituents such as tannins, alkaloids, flavonoids, saponin, glycosides, phenolics, polysachharides, proteoglycans and volatile oils which act against different diseases. Studies showed that the incorporation of medicinal plants in the diet of fish stimulated the immune system of fish and enhance their disease resistance properties. In the present study, the use of some medicinal plants as immunostimulants or phyto-antibacterial agents either injection or dietary administration against fish disease are discussed.*

Keywords*: medicinal plants, immunostimulants, bio-antimicrobials, aquaculture.*

1. INTRODUCTION

Fish farming is the principal form of aquaculture in the world and growing more rapid food animal-producing sector (FAO 2002, 2003). Increasing demands on wild fisheries by commercial fishing operations have caused widespread overfishing. Pisciculture has been employed to compensate the shortage of animal protein all over the world. It has the potential to create huge job opportunities, provided fish cultivation is done on a scientific basis. Disease is the single most important limiting factor in the progressive growth of aquaculture industries. (Sakai, 1999).

The immune systems of aquatic vertebrates, such as those of higher animals, are sensitive

to immune challenges by environmental stresses. High-density fish farming and chemical contaminants in water, especially chronic exposure to toxic pollutants, can lead to decreased resistance to viral, bacterial and parasitic diseases. It has been suggested that the diverse evasive strategies of micro-organisms to their host have acted as inducers of vertebrate defense evolution (Castro *et al.,* 2008).

Fishes are one of the most primitive vertebrates, which possess non-specific defence mechanisms of the invertebrates such as phagocytic mechanisms of macrophages and granular leucocytes. They were also the first animals to develop both cellular and humoral immune responses mediated by lymphocytes. The main lymphoid organs of fish are the thymus, anterior kidney and spleen. In fishes, non-specific immunity is considered as the first line of defence and represents a considerable part of the immune response (Dalmo *et al*., 1997).

Use of antibiotics in fish production increases the prevalence of antibiotic resistance in human diseases. For reducing the usage of antibiotics, immunostimulant is one of the useful tools in aquaculture. Moreover, due to the availability of limited vaccines in few countries and their pathogen-specific protective action, much attention has been directed towards the use of immunostimulants in aquaculture to control infectious diseases. An immunostimulant is a chemical drug, stressor or action that increases the non-specific defence mechanism or the specific immune response. Various herbal products such as from Hygrophila spinosa, Withania somnifera, Zingiber officinale, Solanum trilobatum, Andrographis paniculata, Psoralea corylifolia, Eclipta erecta, Ocimum sanctum, Picrorhiza kurroa, Phyllanthus niruri, Tinospora cordifolia, purified Silajit and cod-liver oil have the characteristics of growth promotion, anti-stress, immunostimulation and anti-bacterial (Sajid *et al*., 2011).

2. SOURCES OF MEDICINAL PLANTS"

The application of natural and innocuous compounds has potential in aquaculture as an alternative to the use of antibiotics. More than 60 different medicinal-plant species have been studied for the improvement of fish health and disease management in aquaculture. Whole or parts of medicinal plants can be used for extracting medicinal compounds. Whole plant *Cynodon dactylonwas* used to prevent the white spot syndrome virus (WSSV) infection in black tiger prawns, while Rosmarinus officinalis was mixed with the feed either as whole dried leaves or as dried ethyl acetate extract (Abutbul *et al.*, 2004).

3. IMMUNOSTIMULANTS

Immunostimulants, also known as immunostimulators, are substances (drugs and nutrients) that stimulate the immune system by inducing activation or increasing activity of any of its components. The immunostimulants could increase the resistance of fish to infectious diseases by enhancing non-specific defence mechanism. Immunostimulants can be administered by injection, bathing or orally, with the latter appearing to be the most practicable (Jeney and Anderson, 1993; Sakai, 1999; Yin *et al.*, 2006). It will boost the potency of the vaccine, thereby decreasing the dose necessary for the same effect (Jeney and Anderson, 1993). To this end, it has been proposed that the use of immunostimulants as a dietary supplement to larval fish could be of considerable benefit in boosting the animal's innate defences with little detriment to the developing animal (Bricknell and Dalmo, 2005).

3.1 Leave extract as immunostimulants

Recently, in aquaculture, scores of plant extracts have been tested and used with good results in the control of bacterial and viral diseases. Fourteen herbs have been tested against *Aeromonas hydrophila* infection in *Oreochromis niloticus*, among them, the ethanol extract of *Psidium guajava* has been found to have highest anti-microbial activity. The stimulation of specific and non-specific immunity and the protection against fish pathogen *A. hydrophila* in *O. mossambicus* by ethanol and petroleum ether extracts of *T. cordifolia* were observed. *Astragalus membranaceus* extract significantly enhanced the phagocytic activity of leucocytes isolated from Nile tilapia (Sakai, 1999).

3.2 Plant parts as immunostimulants

Many parts of the plant materials possess medicinal properties. Numerous plant materials are widely used in aquaculture for preventing diseases by controlling the pathogenic microbes and enhancing the immunity. *A. hydrophila* infection in rainbow trout (*Oncorhyncus mykiss*) was controlled by garlic. Garlic can help in the control of bacteria and fungi and increase the welfare of fish. Several medicinal plant- based materials were administered as immunostimulants in various fish species against pathogens. Natural immunostimulants are biocompatible, biodegradable and safe for both the environment and human health. Moreover, they possess an added nutritional value (Ortuno *et al*., 2002).

3.3 Essential oil as immunostimulants

In aquaculture practice, pathogenic microbes are controlled by essential oils, and they also act as one of the immunostimulants. The carbonyl group of cinnamaldehyde is thought to be responsible for anti-microbial action by binding to cellular proteins and preventing them from functioning properly. Diet supplemented with *Zataria multiflora* essential oil enhanced common carp immunity to some extent even though fish cannot express its potential immunity during low temperature. Some effective components such as thymol and terpinene present in the oils have the properties in stimulating the fish immunity Vaseeharan *et al.*, (2012).

3.4 Herbal drugs as immunostimulators

Papaya leaf meal contains an enzyme, namely papain, which increases the protein digestion, food conversion ratio, specific growth rate and weight gain in 16 % unsoaked papaya meal diet fed to P. monodon post-larvae. Yellowtail fish treated orally with glycyrrhizin showed increased protection against Edwardsiella seriola infection, although lysozyme activity of blood and phagocytic activities of macrophages were not enhanced. The herbal extracts from Astragalus membranaceus, Portulaca oleracea, Flavescent sophora and Azadirachtin, a triterpenoid derived from *A. indica*, enhanced respiratory burst activities, the leucocyte count and the primary and secondary antibody response against SRBC in tilapia (Logambal and Michael, 2001).

Table 1: Main medicinal plant extracts and their multiple therapeutic properties in aquaculture.

English name	Scientific name	Useful part	Active substance	Therapeutic properties
Aromatic species				
Nutmeg	*Myristica flagrans*	Seed	Sabinene	Digestion, stimulant, antidiarrhoic
Cinnamon	*Cinnamomum zeylanicum*	Bark	Ammameldehyde	Appetite and digestion stimulant, antiseptic
Clove	*Syzygium aromaticum*	Cloves	Eugenol	Appetite and digestion stimulant, antiseptic

Caradamon	*Elettaria caramomum*	Seed	Cinook	Appetite and digestion stimulant
Coriander.	*Coriandum sativum L*	Leaves	Unalol	Digestion stimulant
Cumin	*Cuminum cyminum*	Seed	Cuminaldehyde	Digestive, galactagauge
Anise	*Illicum verum*	Fruit	Anethole	Digestive stimulant, galactagauge
Celery	*Apium graveolens*	Fruit,	leaves Phtalides	Appetite and digestion stimulant
Parsley	*Pelroselinum crispum*	Leaves	Apiol	Appetite and digestion stimulant, antiseptic
Fenugreek	*Trigonella foenumgraecum*	Seed	Trigonelline	Appetite stimulant
Pungent species				
Capsicum	*Capsicum annum longum*	Fruit	Capsaicin	Antidiarrhoic, stimulant tonic, antiinflammatory
Pepper	*Piper nigrum*	Fruit	Piperine	Digestion stimulant
Horseradish	*Cochlearia armoracia*	Root	Allyl isothiocyanate	Appetite stimulant
Mustard	*Brassica spp.*	Seed	Allyl isothiocyanate	Digestion stimulant
Ginger	*Zingiber officinale*	Rhizom	Zingerole	Gastric stimulant
Aromatic herbs and spices				
Garlic	*Allium tuberosum*	Bulb	Allicin	Digestion stimulant, antiseptic
Rosemary	*Aniba rosaeodora*	Leaves	Cineole	Digestion stimulant, antiseptic, antioxidant
Thyme	*Thymus vulgaris*	Whole plant	Thymol	Digestion stimulant, antiseptic, antioxidant
Sage	*Salvia apiana*	Leaves	Cineole	Digestion stimulant, antiseptic, carminative
Bay laurel	*Laurus nobilis*	Leaves	Cineole	Appetite and digestion stimulant, antiseptic
Peppermint	*Mentha piperita*	Leaves	Menthol	Appetite and digestion stimulant, antiseptic
Artemisia	*Artemisia annua*	Leaves	Artemisin	Anticoccidial
Neem	*Azadirachta indica*	Leaves, bark	Azadirachtin, salanin, numbin	Antiviral, antiseptic, fungicidal

4. MODE OF ACTION OF MEDICINAL PLANTS"

4.1 Improve the innate immune responses

Several herbal medicines showed an anti-microbial activity, and facilitated growth and maturation of various fish species. Therefore, there is a growing interest in using medicinal herbs as immunostimulants in aquaculture. Many investigations on their effects at molecular mechanism levels have been undertaken, for instance on *Astragalus membranaceus* and *Nelumbo nucifera*. The use of immunostimulants is of an increasing interest for boosting the defense mechanisms and conferring protection of animals from infectious diseases (Sakai, 1999).

4.2 Enhance antimicrobial activity

The antimicrobial properties of medicinal plants and their active compounds have been investigated by many researchers world-wide. Herb extracts have great anti-bacterial activity against both Gram positive and Gram negative bacteria. They can even be used to treat specific diseases caused by virus, parasites and fungi. Indian almond extract was an alternative antibacterial remedy against tilapia ecto-parasites and the bacterial pathogen *A. hydrophila*. *Rosmarinus officinaliswas* used to treat Streptococcus infection in tilapia (*Oreochromis* sp.) (Abutbul *et al.*, 2004).

Table 2: Some important herbs and their anti-microbial properties in aquaculture.

Herbs	Activity	Microorganisms
Garlic	Anti-bacterial	*Salmonella typhymurium, Escherechia coli, Staphylococcus aureus, Bacillus cereus, Bacillus subtilis, Candida albicans*
Onion	Anti-fungal	*Aspergillus flavis,*
Cinnamon	Anti-fungal	*Myctoxigenic aspergillus, Aspergillus parasiticus*
Cloves	Anti-fungal	*Myctoxigenic aspergillus*
Mustard	Anti-fungal	*Myctoxigenic aspergillus*
Allspice	Anti-fungal	*Myctoxigenic aspergillus*
Oregano	Anti-fungal, anti-bacterial	*Myctoxigenic aspergillus, Salmonella* spp.,*Vibrio parahaemolyticus*
Rosemary	Anti-bacterial	*Bacillus cereus, Staphylococcus aureus, Vibrio parahaemolyticus*

Bay leaf	Anti-bacterial	*Clostridium botulinum*
Sage	Anti-bacterial	*Bacillus cereus, Staphylococcus aureus,*
Thyme	Anti-bacterial	*Vibrio parahaemolyticus*

Table 3: Dosage and application of some medicinal plants used to control diseases in aquaculture.

Medicinal plant	Disease	Dosage	Application
Azadirachta indica	Parasite control (Anchor worm, Trichodina, Triprtiella and Trichodinella)	150-200 twigs for a 100 m^2 pond	Immersion
Ricinus communis	Epizootic Ulcerative Syndrome (Red-spot disease)	20-30 kg for a 1000 m^2 in width and 1.5-2.0 m in depth pond	Immersion
Portulacaca oleracea	Bacterial Eanteritis in fecting *Ctenopharyngodon idella*	1.5-3.0 kg in 100 kg^{-1} of fishes	Feeding
Eclipta alba	Parasites	10 g of *Eclipta alba* leaves, 1-3 times day^{-1}	Feeding
Persicaria hydropiper	Endoparasites and Epizootic Ulcerative Syndrome (EUS)	3 kg water-pepper leaves 100 kg^{-1} fishes	Feeding

4.3 Enhance the fish growth, feed utilization and nutrient digestibility

Medicinal plants have been proven as growth promoters. Firstly, they enhance digestive enzymes, and thus boost survival and growth rates of aquatic animals. Three herbs (*Alteranthera sessilis*, *Eclipta alba* and *Cissus quadrangularis*) acted as appetizers and enhanced the activities of digestive enzymes (protease, amylase and lipase) of freshwater prawns. This resulted in an enhancement of food utilization and ultimately in the production of better growth rates as indicated by the evidence of elevated concentrations of vitamins, protein, essential amino acids, unsaturated fatty acids and minerals. (Radhakrishnan *et al.*, 2014).

Table 4: The use of herbal extracts in shrimp boodstock diets.

Botanical Name	Family Botanical	Distribution Botanical	Useful parts Botanical	Biological effect in aquaculture	Reference
Cinnamonum zeylanicum	Lauraceae	India, Sri Lanka	Bark	Endocrine system, Growth promoter	Punitha, 2003
Elettaria cardomomum	Scitaminaceae	India, Burma, Sri Lanka	Dried ripe seeds	Endocrine system, Growth promoter	Punitha, 2003
Eugenia caryophyllata	Myrtaceae	India, Sri Lanka	Fruits and dried flower buds	Endocrine system, Growth promoter	Punitha, 2003
Mesua ferrea	Guttiferae	India, Burma, Andaman, Nicobar Islands	Flowe buds, seeds and bark	Endocrine system, Growth promoter	Punitha, 2003
Asparagus racemous	Liliaceae	India	Leaves and Root	Endocrine system	Devi, 1995
Mucuna pruriens	Papilionaceae	Tropics	Seeds, roots and legumes	Endocrine system	Babu and Marian, 2001
Witania somnifera	Solanaceae	India	Root and leaves	Endocrine system	Babu, 1999; Citrasu, 2008

5. CONCLUSION

Antibiotics, chemotherapeutants and vaccines are expensive and lead to many adverse effects such as bioaccumulation and multi-resistance species development in the environment. Plant materials have a potential application as an immunostimulant in fish culture, primarily because they are not expensive and act against a broad spectrum of pathogens. The preparation of plant extract is much easier and inexpensive. Many plant products are used as anti-bacterial and anti-viral materials. The use of plant products as immunostimulants in fish culture systems may also be of environmental value due to its biodegradability.

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