Marine Ecosystem And Human Health

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I. INTRODUCTION

The oceans are the source of a large group of structurally unique natural products that are mainly accumulated in marine organisms such as sponges, tunicates, bryozoans, molluscs, echinoderms and fishes (Proksch *et al.*, 2002). Moreover the sea is a treasure trove of immense bioactive substances, which are not only a source of potential drugs but also of new and novel structures with useful biological activity. Nevertheless less than 1% of these have been examined for their pharmacological activity.

Marine pharmaceuticals are as old as mankind itself with early fact and fancy art-fully mixed in the use of natural products from the sea. Because of the physical and chemical conditions in the marine environment almost every class of marine organisms exhibit a variety of molecules with unique structural features.

Biologists, chemists, pharmaceutical and medical scientists are taking up the challenge of the drug development from ocean. In the last three decades approximately 2500 new metabolites were reported from marine organisms ranging from prokaryotic microbes and soft-bodied invertebrates to marine mammals. These metabolites are equally diverse in their biosynthetic origins Several bioactive compounds have been isolated from invertebrates like sponges, corals, fishes, sea anemones, crustaceans and mollusks and identified as steroids, terpenoids, isoprenoids, sesquiterpenes. Many bioactive compounds have shown potential of becoming therapeutic agents of near future (Proksch *et al.*, 2002).

Marine natural products continue to be a structurally diverse and pharmacologically most interesting source of bioactive metabolites. Some of them hold great potential for the development of new and much needed drugs primarily in the treatment of cancer.

In the recent past, several pharmacological substances of marine origin have been developed. The “Cephalothin” (marketed as Keflin by Lilly pharmaceutical Co. USA) is an antibiotic, active against a number of penicillin resistant bacteria .The neurotoxin isolated from the porcupine fish and puffer fish have been used clinically as a drug in terminal cancer and successful isolation of compounds such as protamine, cephalosporin C, Ara-A and Ara-C are a few examples.

Many marine bioactive compounds have shown potential of becoming therapeutic agents in near future viz. Chitosan as an anti-obesity agent (Singla *et al.*, 2001), Didemin B, Aplidineand ET-743 as antitumor agents (Rinehart, 2000), Manoalide and Ziconotide as an analgesic (Olivera, 2002) and Kahalalides against HIV Ziconotide is the synthetic equivalent of a neuroactive peptide found in the venom of the fish-hunting marine snail Conus magus, its analgesic efficacy has been demonstrated in both animal and human studies (Eldabe *et al.*, 2007). The ether extract of ray fishes showed very significant inhibitory effect against *Vibrio cholerae* in comparison with known antibiotic-tetracycline (Parabhdeep, 2009).

Fish constitute almost half the number of vertebrates on earth and approximately 31000 species of fishes are contained in some 50 orders and 445 families (Nelson, 2006). Fishes, like many other forms of life are of immense value to human beings in various ways. They have been a staple food item in the diet for many people and they also serve as a curative source of various ailments and vitamin deficient diseases.

Recent discovery suggest that the mucus produced by some fish could be used to make a new sunscreen. Fish that live around coral reefs have been found to have chemicals called Mycosporine like amino acids in their mucus. These chemicals block ultraviolet light. The mucus of a fish provides physical protection by trapping pathogens. When the old mucus layer containing the pathogens is shed and replaced by new mucus, the pathogens are lost. Antibodies, antimicrobial peptides and enzymes in the mucus actively attack pathogens. Antibodies, antimicrobial peptides and enzymes in the mucus actively attack pathogens (Crampton, 2017).

Therefore fishes still represents source of pharmacological compounds that may be useful as research tools or lead compounds for drugs, and as such, there pharmacological actions have been the focus of research work.

II. IMMUNOLOGY OF FISH

The immune system is composed of numerous organs and cells that act together in a dynamic network in the defense against infection, disease and foreign substances. However, under normal conditions the fish maintains a healthy state by defending itself against these potential invaders by a complex system of innate defense mechanisms. The vertebrate immune system is composed of two types of immunity, innate and adaptive.

Adaptive immunity emerged early in vertebrate evolution, at some time during the division of the jawless lamprey and the cartilaginous fishes (Pham, 2008). Innate immunity developed before the divergence of vertebrates and invertebrates, and most multicellular organisms (e.g. invertebrates) depend on it completely (Khan, *et al*., 2000). In all vertebrates, initial penetration by microorganisms into the body is firstly encountered by innate defense mechanisms, making innate immunity a pivotal barricade innate host defenses are defeated or by passed and when the elimination of a new infection is unsuccessful. Innate mechanisms are both constitutive and responsive (i.e. existing or inducible) and provide protection by preventing the attachment, invasion or multiplication of microbes on or in the tissues.

Many of the innate immune mechanisms of higher vertebrates have been identified in fish. Immune responses in ectothermic vertebrates display many analogies to those of higher vertebrates (Mohammad, 2016).

III. MUCUS AS AN IMMUNOLOGICAL FACTOR IN FISH

The skin of fish is a dynamic tissue whose cellular make up is known to be influenced by factors such as season, stress, diseases, development stage and environmental conditions.A great deal of research supports the notion that layers of mucus accumulate on the skin and gills of fish that are stressed by disease, adverse environmental conditions and handling (Ristow and Schmeisser, 2008). The mucus protects the skin from pathogens and suspended particles and its alarm substance mucin has potential of antimicrobial and noxious properties (Ryan, *et al.,* 2011).

Mucus plays an important role in the prevention of colonization of parasites, bacteria and fungi (Sakat, *et al.,* 2010). The functional property of the mucus depends on its capacity to form gel on the epithelial surface (Bragadeeswam *et al*., 2011). This mucus is secreted by the epidermal goblet cells, composed mainly of water and gel forming macromolecules such as mucins and other glycoproteins (Martinez *et al*., 2006).

Mucins are strongly adhesive, play a major role in the defence of the mucosae form a matrix in which a diverse range of antimicrobial molecules can be found and impart viscoelastic and rheological properties to mucosal layers (Thornton *et al*., 2008). The mucus produced by the epidermal goblet cells of this species is rich in mannose, N-acetylgalactosamine, and N-acetylglucosamine residues (Uzzell *et al*., 2010).

In addition to AMP, fish mucus also contains a variety of biologically active substances such as lysozyme, lectins, flavoenzymes, and immunoglobulin. It was reported that epithelial tissues produce antimicrobial molecules which serve as the first line of a host's defense against microbial invasion in a variety of vertebrates (Ashwin, 2015).

Mucus membranes lining the alimentary, respiratory, and urogenital tracts are equipped with a layer of mucus which functions to entrap foreign microorganisms out of the body.

The mucus layer is suggested to be multifunctional by displaying traits and actions important in e.g: Osmoregulation, reduction of friction and diseases resistance (Villarroel, *et al.,* 2007).

The skin surfaces support considerable concentration of gradients, particularly for sodium and chloride in fresh water or seawater. Fish biologists suspected that mucus might be involved in ion regulation. The abundance of goblet cells on fish surface may also be correlated with environmental salinity (Wang, *et al.,* 2010) and this has contributed to the view that mucus somehow supports ion regulation by fish. In general the abundance of goblet cells on gills and non-gill surface decreases as salinity increases .Antibacterial activity in mucus has been demonstrated in several fish species (Oren, 2008) yet this activity seems to vary from fish species to fish species and can be specific towards certain bacteria (Park *et al.,* 1998).

Mucus and osmoregulation in fish

Mucus is helpful for a fish because in conjunction with the scales it partially blocks the movement of water into and out of the fish's body. This helps to maintain constant conditions inside the fish.

Other parts of the body also influence the salt and water concentration in the fish. The urine contains more or less water and salt, as necessary. In addition, the gills excrete or absorb salts, depending on a fish's needs once again, and the arrows into and out of the skin are short due to the presence of scales and mucus.

IV. PEPTIDES

Peptides play an important role as mediators of key biological functions, their unique properties, in turns of efficacy, selectivity, specificity, low toxicity make them particularly attractive as therapeutic, prophylactic diagnostic reagents for diverse indications such as allergy, cardio vascular disease, infectious disease, immunological disorder, gastro intestinal dysfunction, cancer and imaging and The protein therapy can be ambiguous. Since any polymer of two or more amino acids linked by peptide bonds is considered a peptide but the minimum number of amino acid residues required to form a protein is arbitrary. Here we define a cut of value is hundred, so that any change fewer than 100 amino acids considered a peptide and not a protein (Lien, 2014).

Peptides were initially considered poor drug candidates because of their inability to cross biological membrane and cellular barrier efficiently, and in some cases also there low intrinsic stability reflecting physicochemical properties such as their solubility in water and their susceptibility to degradation following pH changes. Indeed, naked peptides are degraded by proteolytic enzymes in gastro intestinal tract and plasma and are rapidly removed from circulation by hepatic and renal clearance (Vlieghe *et al.*, 2010).

Although intrinsic peptides in stability are detrimental and some therapeutic applications, it is beneficial when peptides are conjugated to cancer drugs for chemotherapy as these prevents the accumulation of toxic molecules in the body (Ready *et al.*, 2011).

However, because of their diverse pharmacological properties to improve the pharmacokinetics properties of peptides includes the following

- Modification of terminal amino acids (example by N-terminal acetylation or C- terminal amidation and substitution of residues at predicted cleavage sites with non-natural counterparts such as D-amino acids.
- Circularization or peptide bond modification to enhance bioavailability and selectivity.
- Conjugation with polyethylene glycol (PEG) to reduce renal and hepatic clearance, or encapsulation within liposomes or similar nanoparticles based on polylactic or polylactic glycolic acid to achieve targeting specificity and controlled release.

These technological advances in peptide synthesis have encouraged the pharmaceutical industry to invest in the development of novel peptide-based drugs, focussing on efficient alternatives to parental administration, which is one of the major obstacles to their wide spread adoption.

Hundreds of peptides produced by either chemical synthesis or biosynthesis in recombinant cells have already reached the market are in advanced clinical trials (Vlieghe *et al.*, 2010). Most peptides are hormones, receptor agonists or antagonists, enzyme inhibitors and diagnostic reagents, but in the near feature it is likely that vaccines Antimicrobial Peptides (AMPs) are ubiquitous in the living kingdom.

IV. ANTIMICROBIAL PEPTIDES [AMPS]

Antimicrobial peptides (AMPs) are increasingly recognized as a critical component of the host's defence against infection. AMPs are antibiotics that have been isolated from a multitude of organisms ranging from microbes to plant and animal species. The AMPs show variations in their biochemical properties such as amino acid sequences, length, and structure, yet they share several common features. They display a broad spectrum of activity against numerous pathogenic organisms including Gram-positive and Gram-negative bacteria, yeast, fungi, enveloped viruses, and parasites with little or no toxicity to host cells.

These AMPs are present in tissues exposed to microorganisms such as mucosal surfaces and skin and immune cells such as mast cells. AMPs are produced constitutively or induced upon infection in fish epidermal mucus to defend against invading pathogens.

Several types of AMPs have been identified from mucosal tissues or immune cells of a number of teleosts, and they are, at present, considered a very important part of the mucus and skin barrier function. Alpha-helical amphipathic peptides are very common in fish, and they have been recently reviewed.
AMPs possess other desirable characteristics which may be exploited in the near future as antimicrobial agents, vaccine adjuvants, inactivated vaccines, and antitumor agents.

AMP History

The oldest but also most famous antimicrobial peptide, nisin, was first described in 1928. This 34-residue peptide is produced by a strain of lactic acid bacteria, *Lactoc occuslactis*. It belongs to the antibiotics, a class of highly post-translationally modified bacteriocins. Its structure was elucidated in 1971.Melittin is the main peptide component of the venom of the European honey bee, *Apis mellifera*. These two ancient peptides represent the great diversity of AMPs. Indeed, these gene-encoded peptides are light-years away at structural and activity levels. In fact, the first, highly modified, presents a narrow spectrum of activity while the second, unmodified, is active against prokaryotic and eukaryotic cells. This class of antibiotics was re-discovered at the end of this decade with widespread antibiotics bioprospecting giving birth to a "second generation" of AMPs.

General properties of AMPS

AMPs have been shown to be ubiquitous and to be produced by organisms ranging from bacteria to superior mammals. Ribosomally synthetized AMPs from prokaryotes are called bacteriocins. They have been demonstrated to be potent weapons to defend or to capture an ecological niche (Ristow, 2008). In Eukaryotes, antimicrobial peptides like thionins have been described in various organs of plants (Ashwin, 2015). Nevertheless most AMPs have been investigated in animals. In insects and amphibians, a multitude of AMPs have been described. They are also produced by bird's and mammals. In a nutshell, during the last quarter of a century, a plethora of ribosomally synthetized AMPs have been described and characterized.

Most AMPs are 20 to 60-residue long, cationic and amphipathic peptides. Based on a 1997 peptide sequence database, statistical data reveal that the average AMP is 30.5 residues long and exhibits a cationic net charge, + 3.1. Of course, exceptions exist in terms of size or charge (Wang *et al.*, 2007). Overall, two main categories can be distinguished i.e. linear peptides and disulfide bridge-containing peptides. A global classification based on structural homology is now accepted.

Studies have shown that the same antimicrobial peptide may act on different viruses through different mechanisms.

Fish AMP'S

Unlike invertebrates which depend on innate effectors for warding off microbial invasion, adaptative immunity is present in vertebrates. In fish in particular, the acquired immune response is apathetic due to a limited antibody panel and slow lymphocyte proliferation
AMPs, one of the main components of fish innate immunity, play a crucial part in fish defence. Moreover, various peptides have been characterized such as pardaxin from *Pardachirus sp.*, Misgurnin from *Misgurnus sp.* (Park *et al.*, 1997) and Moronecidin from *Morone sp.*

Classification of AMPS

Based on both structural features and origins, fish AMPs are classified into four classes:

- AMPs exhibiting an α- helical structure (pleurocidins, moronecidins and piscidins)
- β-sheet structured AMPs containing 4 disulfide bonds (hepcidins)
- Proteolytic fragments of structural or functional proteins
- AMPs produced by fish-associated bacteria

Mechanisms of action of antimicrobial peptides

Common mechanisms of action of antimicrobial peptides on bacteria, declined in membrane permeabilization processes and intracellular effects (Nguyen *et al.*, 2011). When reaching a threshold concentration, the peptides at the outer surface of the membrane can insert themselves into the membrane.

1. Form peptide- lined pores according to the barrel-stave model.
2. Create micellar structures with the membrane by interacting with its lipids, in the carpet model.
3. Form peptide-and-lipid-lines pores according to the toroidal pore model. In comparison, the discordered toroidal model involves less peptide.
4. The coming of peptides in between the membrane can increase its thickness, sometimes generating a non- bilayer intermediate form.
5. All these mechanisms of action can occur altogether for the same molecule. In the cytoplasm, various disturbances can be generated by AMPs, from DNA replication to cell-wall synthesis. Buforin II, pleurocidin and dermaseptin can inhibit DNA and RNA synthesis.
6. Indolicidin and PR-39 are known to limit protein synthesis in targeted bacterial cells
7. Other kinds of enzymatic activity can also be altered by AMPs. Pyrrhocidin limits the ATPase activity DnaK, which is implied in the chaperone-assisted protein folding.

8. The enzymatic modification of aminoglycosydes has also been shown to be inhibited by AMPs.
9. Finally, lantibiotics such as the well-known niscin and mersacidin have been demonstrated to disturb the synthesis of the cell-wall molecules.
10. Among other structural components.

Abbreviations:

LEAP: liver-expressed antimicrobial peptide, HAP: hematopoietic antimicrobial peptide, CRP: Corticostatin-related peptide. Antimicrobial differs from Antibacterial by fungal activity.

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