An Update of Terpenoids, Steroids and Biodiversity of Seaweeds from the Coasts of Pakistan

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(Received on 26th September 2009, accepted in revised form 12th January 2010)

Summary: This review article presents the comprehensive update of the ecology, biodiversity and importance of seaweeds. In addition, the article also presents the terpenoidal and steroidal compounds isolated from seaweeds of the coastline of Pakistan during the period of 1980 to 2008. Rapid growth in this field during the last thirty years resulted in the identification of a surprisingly large number of new structures of terpenoids and sterols. Overall, thirty-two terpenoids and eighteen steroids have been reported from the seaweeds of the coasts of Pakistan. The main sources of these compounds are brown, red and green algae. Furthermore, the study of the literature showed that only few terpenoids and steroids have been investigated for bioassay screening which exhibited some kind of activities to a little extent.

Introduction

The oceans and seas which cover two-third of the total area of the globe, have served as a source of food, transportation, bio-fuels, fertilizers and medicines [1]. The main sources of bioactive marine natural products are fish, crabs, mollusks, tunicates, sponges and different kinds of algae [1]. As a result of the potential for new drug discovery from marine bio-sphere, marine natural products have attracted scientists from different disciplines such as organic chemistry, bioorganic chemistry, pharmacology, biology and ecology. This interest has led to the discovery of thousands of marine natural products and many of the compounds have shown very promising biological activities [1-6]. A number of bioactive compounds isolated from marine invertebrates such as sponges, coelenterates, gorgonians, soft and hard corals, algae, mollusks etc. [7]. Stypopodium, a potent cytotoxin produced by Stypopodium zonale, appears to inhibit cancer cell damage [7]. Manolide, a secondary metabolite belongs to sesterpene isolated from Luffariella variabilis sponge has anti-inflammatory and analgesic properties in addition to its anti-leukemic and anti-fungal properties [8]. The antiviral drugs Ara-A, Zidovudine (AZT) and the anti-cancer agent Ara-C, developed from extracts of sponges found on a caribbean reef were among the earliest modern medicines obtained from coral reefs [8]. AZT was the first drug approved for the treatment of AIDS and HIV infection [8]. Coral reefs are storehouses of genetic resources with vast medicinal potential but they must be properly managed [8]. Squalamine is a strong naturally-derived broad-spectrum antibiotic that is predominantly derived from the livers of dogfish and other shark species [9]. Didemnin B was the first defined marine natural product to enter clinical trials as a potential anti-cancer drug [9, 10]. Kahalalide F was isolated from a Sacoglossan (sea slug) mollusk, Elysia rufescens collected from Hawaii [11]. The drug shows promise in treating a broad range of tumors, including non-small cell lung cancer (NSCLC), melanoma, androgen-independent prostate cancer and hepatocellular carcinoma [11]. More recently, mazamine A was shown to have potent anti-malarial activity in an assay against rodent malaria parasite Plasmodium berghei in vivo [12, 13].

In Pakistan, the plant parasitic nematode problem has been recognized as one of the major threats to the agricultural production [14]. The use of chemical nematicides for the control of plant parasitic nematodes is a common practice but it is an expensive and risky operation [14]. Many researchers have studied marine organisms as a source of bioactive compounds showing antiviral, antifungal, antibacterial and nematicidal activities [14]. Stoechospermum marginatum (seaweed) constitutes a prodigious source of new and structurally complex secondary metabolites, has shown promising results in the control of Meloidogyne javanica root-knot nematode [14]. The literature survey stimulated us
that there is an urgent need to further explore the bioactive principles of marine organisms.

Algae

The simplest plants found in the plant kingdom belong to the group known as algae. The algae differ from higher plants because they do not possess true roots, stems or leaves. According to Trainor “algae are photosynthetic and nonvascular plants that contain chlorophyll and have simple reproductive structures” [1, 15]. The written record of the study of marine plants begins in the third century B. C. with the Greek naturalist Theophrastus who gave descriptive accounts of seaweeds and grouped them along the terrestrial plants in his classification of trees, shrubs and herbs. The seaweeds received no considerable attention for about 1800 years. The elaborately illustrated work of Gmelin evidently generated considerable interest in the collection and preparation of seaweed specimens. The algal herbarium began to be assembled all over the Europe [16]. Generally, different classes of algae are distributed around the globe such as China, Europe, America, Central Pacific, Hawaii, Japan, Indonesia, Australia, Caribbean, Pakistan, New Zealand, South Africa, India and Bangladesh etc [1, 15]. Algae play an important role in the direct economy of many countries. Seaweeds have significant commercial applications as vegetables, animal fodder, fertilizer, bio-fuels and as a major source of medicines for the cure of biological disorders. The well known commercial products obtained from seaweeds are, chlorellin (antibiotic) from Chlorella; agar and alginic acid from Laminaria and Macrocytis pyriforma [1, 15]. On the basis of number of cells and the size of the algae they are mainly classified into two groups namely microalgae and macroalgae which are described as bellow:

Microalgae

Microscopic algae are called microalgae that live as single cell or colonies. The earth’s surface is composed of 70% sea water which contains a wide range of unicellular plants known as microalgae. Four most important classes of microalgae in terms of abundance are the Bacillariophyceae (diatoms), the Chlorophyceae (green algae), Chrysophyceae (golden algae) and Cyanophyceae (blue-green algae/Cyanobacteria) (Tables-1, 2). Diatoms belong to the class Bacillariophyceae are often regarded as the most beautiful of the algae. Each diatom has a cell wall made of glass that is very finely etched with a species-specific pattern of dots and lines. The patterns on the diatom cell walls are so precise that they were used for years to test the optics of new microscopes. Diatoms are also the most abundant algae in the open ocean and responsible for about one-quarter of all the oxygen gas produced on the earth each year. Diatom’s populations often bloom in lakes in the spring, providing a major food for zooplankton, forming the base of the aquatic food chain. There are over one hundred thousand species of diatoms.

Green algae belong to the class Chlorophyceae that may be unicellular, multicellular (seaweeds) and colonial. Most of the green algae are aquatic and are found commonly in freshwater (mainly Charophytes) and marine habitats (mainly chlorophytes); some are terrestrial, growing on soil, trees or rocks. Green algae are closely related to plants. They have the same pigments (chlorophyll a, b and carotenoids), chemicals in their cell walls (cellulose) and the same storage product (starch) as plants. There are over 8,000 species of green algae found in the nature but only 20% are marine algae. Green seaweeds are also small, with a similar size range to the red seaweeds (Tables-1, 2).

Golden algae belong to the class of Chrysophyceae that are a large group of algae which are formally known as Chrysophytes. There are more than 1000 species of golden algae found in both marine and fresh waters. They are characterized by the pigment fucoxanthin and oil droplets as the food-reserve. A majority of Chrysophytes are unicellular and free-swimming, colonial and filamentous. In many lakes, golden algae are the primary source of food for zooplankton and they can therefore be extremely important for the entire ecosystem of a lake. Some species of Chrysophytes are colourless, but the vast majority is capable of carrying out photosynthesis. They are known to feed on diatoms and bacteria (Tables-1, 2).

Cyanobacteria are microalgae belonging to the class of Cyanophyceae which are microscopic in size, often unicellular and are best known by the blue-green algae. The first algae appeared more than three billion years ago were the Cyanobacteria. From these cells, more complex cells developed one and a half billion years later. Today, they have evolved into
Table-1: Classification of algae.

<table>
<thead>
<tr>
<th>Kingdom</th>
<th>Domain</th>
<th>Group</th>
<th>Phylum/Division</th>
<th>Class</th>
<th>Types of algae</th>
<th>Pigmentation</th>
<th>Microalgae/Macroalgae**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protista</td>
<td>Eukaryotes</td>
<td>Archaeplastida</td>
<td>Chlorophyta</td>
<td>Chlorophyceae</td>
<td>Green algae</td>
<td>Microalgae</td>
<td>(unicellular)</td>
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<td></td>
<td></td>
<td>Rhodophyta</td>
<td></td>
<td>Rhodophyceae</td>
<td>Red algae</td>
<td>Microalgae</td>
<td>(unicellular)</td>
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<tr>
<td></td>
<td></td>
<td>Glaucophyta</td>
<td></td>
<td>Glaucophyceae</td>
<td>Red algae and Viridiplantae*</td>
<td>Microalgae</td>
<td>(unicellular)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>Bacillariophyceae</td>
<td>(diatoms)</td>
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<td>Microalgae</td>
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<td></td>
<td></td>
<td>Chromista</td>
<td></td>
<td>Actinophyceae</td>
<td>(Axonina)</td>
<td>Microalgae</td>
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<td></td>
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<td>(Chromalveolata),</td>
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<td>Microalgae</td>
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<td>Alveolata</td>
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<td>Microalgae</td>
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<td>Heterokonta</td>
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<td>Microalgae</td>
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<td></td>
<td>Phaeophyceae</td>
<td>Brown algae</td>
<td>Microalgae</td>
<td>(multicellular)</td>
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<td></td>
<td></td>
<td>Chrysophyceae</td>
<td>Golden algae</td>
<td>Microalgae</td>
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<td>Microalgae</td>
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<td>Microalgae</td>
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<td></td>
<td></td>
<td>Microalgae</td>
<td>(unicellular)</td>
</tr>
</tbody>
</table>

**Viridiplantae: Green algae and land plants; **Macroalgae are also called seaweeds.

thousands of different species of photosynthetic organisms and have become important producers in the oceans, oxygenating the waters in their immediate vicinity by more or less acting as the lungs of the aquatic systems (Tables-1, 2).

**Macroalgae (Seaweeds)**

Macroalgae or “seaweeds” are multicellular and fast growing plants that can reach sizes up to 60 m in length. There are fifteen classes of algae but only three are present in sufficient quantities in nature and have direct commercial importance which include Phaeophyceae (brown algae), Rhodophyceae (red algae) and Chlorophyceae (green algae) (Khan, 2000) (Table-1, 2). The brown algae are a large group of mostly marine multicellular algae. Worldwide there are about 1500-2000 species of brown algae and the majority of these species are found in cold temperate to arctic marine water [17]. The main food reserve of brown algae is the polysaccharide laminarin. Brown seaweeds are usually small (30-60 cm long), medium (2-4 m long) and large enough (up to 60 m long) (Tables-1, 2).

Red Algae belong to the class of Rhodophyceae are mostly marine in origin and include many edible and economically important species. There are 4000-6000 species of red algae which are abundant in the tropical and subtropical
regions of the oceans. Only less than 2% of the red algae are found in freshwater [16]. There are certain types of algae which actually play a pivotal role in creating reefs. These types of red algae are known as coralline algae that built up a layer of carbonate to form a reef structure. Red algae are also the source of carrageenan and agar which are used as food thickeners and stabilizers. Many types of red algae are also used as food throughout the world, especially in Asia. They are usually smaller, generally ranging from a few cm to about a metre in length. Red seaweeds are not always red they are sometimes purple or brownish red. Red colour of these algae is due to the presence of the pigment phycoerythrin which reflects red light and absorbs blue light (Tables 1, 2).

Ecology and Biodiversity

Seaweeds are a fascinating and diverse group of organisms living in the earth's oceans. Some of the seaweeds are attached to rocks in the intertidal zone, some form underwater forests and some are floating on the ocean’s surface. They can be very tiny or quite large growing up to 60 m long. Chlorophyll is also responsible for the green coloration of seaweeds. In addition to chlorophyll some seaweed contains other light absorbing pigments. These pigments can be red, blue and brown or golden which are responsible for the beautiful coloration of the algae [18-20]. Instead of roots, seaweeds have holdfasts, which attach them to the sea floor. A holdfast is not necessary for water and nutrient uptake, but is needed as an anchor. Holdfasts are made up of many fingerlike projections called haptera. The stalk or stem of seaweeds is called a stipe. The function of the stipe is to support the rest of the plant. The structure of the stipe varies among seaweeds which can be flexible, stiff, solid, gas-filled, very long and short. The leaves of seaweeds are called blades that provide a large surface for the absorption of sunlight and in some species the blades also support the reproductive structures of the seaweeds. Some seaweed has only one blade, which
may be divided, while other species have numerous blades. A number of seaweeds have hollow, gas-filled structures called floats or pneumatocysts. The term thallus refers to the entire plant body of seaweeds [18-20].

The Arabian sea Large Marine Ecosystem (ASLME) is characterized by its tropical climate. It encompasses three sub-regions in the Indian Ocean. The Western Arabian sea borders Somalia, Yemen and Oman; the Central Arabian sea borders Iran, the Eastern Arabian sea borders India and Pakistan. Each sub-region has its own originality in terms of current patterns, physical characteristics, physiochemical qualities, dominant species and biodiversity. The seaweeds may be regarded as forming three great zones so far as their usefulness is concerned. There is the north temperate and cold water zone in which large brown seaweeds occur in great quantity. There is central warm water zone in which the principle valuable seaweeds belong to the Rhodophyceae, and there is south temperate and cold water zone where again big areas of brown seaweeds are to be found with considerable quantities of red algae [21].

Marine area of Karachi, the largest city of Pakistan has a coastline of about 100 km at the Northern boundary of Arabian sea. There is a luxuriant growth of seaweeds and invertebrates and on the coastline of Arabian sea [1, 15]. They possess a broad spectrum of application in medicines, fertilizers, fuels, animal fodders and vegetables (Table-3) [1, 15]. These organisms are the major source of biactive compounds which can be used for the cure of different biological disorders [1-6]. The concentration of nutrients in the upper layers of sea water is attributed to the freshwater influx from Indus River or due to upwelling process from bottom layers. The discharge from Indus contains nutrient rich water. In the ocean, most of the nutrients are released soon after an organism dies and are quickly recycled in the waters of aphotic zone. These deep waters emerge by upwelling along the coast as well as in the off shore areas. Thus nutrient rich waters are available within the limit of euphotic zone. These deep environmental conditions create high primary production in the upper layers of Northern Arabian sea. Several other oceanographic factors contribute to the productivity of seaweeds [22]. Northern Arabian sea has a strong thermal stratification near its surface in the upper 200 m. This barrier creates partial inhibition in the vertical mixing of nutrients. Oceanic processes which prevail from time to time in the Arabian sea, such as wind induced upwelling, divergence at current boundaries, internal waves or simply mixing of water by strong wind can shift upward the nutrients to create conditions favorable for high productivity of seaweeds [22]. The oxygen concentration in the Arabian sea drops sharply immediately below the surface. On many occasions the oxygen minimum occurs at the depths less than 50 m and this high oxygenated layer is continued to the surface. The formation of oxygen minimum is mostly due to high production in the euphotic zone, sinking of large amount of organic matter and strong thermal stratification in upper 200 meters, which reduces the exchange of oxygen between the atmosphere and water surface. The high level of nutrient and dissolved oxygen proximity to the surface are conducive for a high productivity of seaweeds in the Arabian sea [22].

The green seaweeds occupy a large area on our shores and show great variation in type and species. During specific season the entire rocky platforms and ledges appear to be covered by a beautiful green carpet due to the presence of green algal vegetation, which mostly comprises of the genus Ulva (also called sea lettuce) and shows great diversity in type and species on our coast. Similarly, another green algal genus Codium forms spongy bushes all along the shores of the sandy beaches, where it is brought as drift material after being broken by upcoming waves [23]. Seaweeds are affected by the physical and chemical changes in their surrounding environment because the growth of the seaweeds is mainly based on the nutrients of the surrounding water. The constant motion of the seas and ocean waves also subjects seaweeds to mechanical stress. Most of the seaweeds that live in the rocky intertidal communities are subjected to the stresses associated with exposure to air and weather conditions. They also face major changes in the temperature and salinity due to water evaporation process. Some seaweeds can dry out almost completely when the tide is out, then take up water and fully recover when the tide brings water back to them. Seaweeds living in tidepools are exposed to changes in water temperature and salinity caused by weather conditions. On hot, sunny days the water in tidepools warms up and evaporates, which increases the salinity of the water. In the rainy days an opposite effect takes place on the seaweeds and during cold days seaweeds are damaged due to freezing [18-20].
Table 3: Comparison of seaweeds from the coasts of Pakistan.

<table>
<thead>
<tr>
<th>Seaweeds</th>
<th>Phylum/Division</th>
<th>Class</th>
<th>Family</th>
<th>Type of algae</th>
<th>Pigmentation</th>
<th>Reported composition</th>
<th>Geographical distribution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nostocystis (=Cystoseira indica)</td>
<td>Heterokonts</td>
<td>Phaeophyceae</td>
<td>Cystoseiraceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin and carotenoids</td>
<td>Steroids, terpenoids, fatty acids, and aromatic compounds</td>
<td>Pakistan and India only</td>
</tr>
<tr>
<td>Dictyota dichotoma</td>
<td>Heterokonts</td>
<td>Dictyotaceae</td>
<td>Dictyotaceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin and carotenoids</td>
<td>Terpenoids, fatty acids and other compounds</td>
<td>India, Pakistan, Philippines, Japan, Australia, Adriatic Ocean, West Indies, Florida, England, Spain, France, Italy, Yugoslavia, Greece, Libya and Morocco</td>
</tr>
<tr>
<td>Jettia lacentifolia</td>
<td>Heterokonts</td>
<td>Phaeophyceae</td>
<td>Siphonophoraceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin</td>
<td>Steroids, fatty acids and aromatic compounds</td>
<td>Pakistan, Oman, Yemen and Brazil</td>
</tr>
<tr>
<td>Sphacelaria variablea</td>
<td>Heterokonts</td>
<td>Phaeophyceae</td>
<td>Dictyotaceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin</td>
<td>Steroids, aromatic compounds, fatty acids, and other compounds</td>
<td>Pakistan, India, Indonesia, Somalia and Yemen</td>
</tr>
<tr>
<td>Dictyota indica</td>
<td>Heterokonts</td>
<td>Phaeophyceae</td>
<td>Dictyotaceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin</td>
<td>Terpenoids, fatty acids and other compounds</td>
<td>Pakistan, India and some other countries</td>
</tr>
<tr>
<td>Fucus vesiculosus</td>
<td>Phaeophyceae</td>
<td>Siphonophoraceae</td>
<td>Macroalgae (brown algae)</td>
<td>Chlorophyll and brown pigment fucoxanthin</td>
<td>Steroids, terpenoids, aromatic compounds, fatty acids, amino acids, carbohydrates, proteins and other compounds</td>
<td>Pakistan, India, South Africa and other countries</td>
<td></td>
</tr>
<tr>
<td>Laurencia pinnatifida (=Pepper dulse)</td>
<td>Rhodophyta</td>
<td>Rhodophyceae (Florideophyceae)</td>
<td>Rhodophyta</td>
<td>Macroalgae (red alga)</td>
<td>Chlorophyll and red pigment phycobilins or phycerythrin</td>
<td>Terpenoids, fatty acids and other compounds</td>
<td>Pakistan, India, Atlantic Island, Australia, New Zealand and other countries</td>
</tr>
<tr>
<td>Laurencia obtusa</td>
<td>Rhodophyta</td>
<td>Rhodophyceae</td>
<td>Rhodophyta</td>
<td>Macroalgae (red alga)</td>
<td>Chlorophyll and red pigment phycobilins or phycerythrin</td>
<td>Steroids, fatty acids and other compounds</td>
<td>Pakistan, India and some other countries</td>
</tr>
<tr>
<td>Seaweeds</td>
<td>Phylum/Division</td>
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<td>Type of algae</td>
<td>Pigmentation</td>
<td>Reported composition</td>
<td>Geographical distribution</td>
</tr>
<tr>
<td>Meehania nodosa</td>
<td>Rhodophyta</td>
<td>Rhodophyceae</td>
<td>Rhodophyta</td>
<td>Macroalgae (red alga)</td>
<td>Contains chlorophyll and red pigment phycobilins or phycerythrin</td>
<td>Steroids, aromatic compounds, fatty acids, and other compounds</td>
<td>Pakistan, Americas, British Columbia and Oregon</td>
</tr>
<tr>
<td>Codium decortication</td>
<td>Chlorophyta</td>
<td>Bryopsidophyceae</td>
<td>Codiaceae</td>
<td>Macroalgae (green algae)</td>
<td>Posses photosynthetic pigments chlorophyll a and b, α- and β-carotene, and xanthophylls</td>
<td>Steroids, fatty acids and other compounds</td>
<td>Pakistan, India, Estonia, Greece, Italy, Portugal, Spain, Turkey, Atlantic Island, America, Caribbean Island, Antarctic, Subantarctic Island and Pacific Islands</td>
</tr>
<tr>
<td>Codium spenceri</td>
<td>Chlorophyta</td>
<td>Bryopsidophyceae</td>
<td>Codiaceae</td>
<td>Macroalgae (green algae)</td>
<td>Posses the photosynthetic pigments chlorophyll a and b, chlorophyll b, and xanthophylls</td>
<td>Steroids, fatty acids and other compounds</td>
<td>Pakistan and India only</td>
</tr>
<tr>
<td>Valonia radiata</td>
<td>Chlorophyta</td>
<td>Ulvophyceae</td>
<td>Valoniaceae</td>
<td>Macroalgae (green algae)</td>
<td>Posses the photosynthetic pigments chlorophyll a and b, chlorophyll b, and xanthophylls</td>
<td>Terpenoids, fatty acids and other compounds</td>
<td>Pakistan, India, Kenya, Madagascar, Mauritius, Mozambique, Reunion, Seychelles, Senegal, South Africa and Tanzania</td>
</tr>
</tbody>
</table>

Seaweed's life and reproductive cycles are quite complicated i.e. some seaweed lives for many years, while others live for only one year. Annual seaweeds generally begin to grow in the spring and continue throughout the summer. During powerful fall and winter storms the stipes and blades of seaweeds are often ripped off. If the holdfast manages to survive through the winter, new blades will begin to grow from it in the spring [18-20].

Importance of Algae

Seaweeds play an important role in the economic growth of various countries because seaweeds are used as nutritional food supplements, alternative health and to provide minerals and vitamins. In addition, they are used to produce a number of commercial products including medicines, agar, alginate, carrageenan, biofuels etc. Algae are becoming a great biotic indicator of environmental changes and human induced alterations. In addition, algae are also used for the wastewater treatment. On the basis of the gradual increase in the demand, the farming of seaweeds has expanded rapidly therefore, a number of countries across the world commercially harvesting the seaweeds on routine basis [1, 16, 21-23].
Medicines

Seaweeds are used to treat or prevent goiter, glandular troubles, stomach disorders, intestinal and bladder difficulties, unusually profuse menstrual flow, high-blood pressure and high plasma-cholesterol level. The *Gracilaria* species are used locally as pain relievers and ointments. Seaweeds may have curative properties for tuberculosis, arthritis, colds and influenza (or flu), worm infestations and even tumors. Some studies have found that seaweeds promote weight loss. For this reason, seaweed extracts is used in some diet pills [1, 16, 21-23]. The crude extracts of Kombu (Laminaria and Ecklonia), Haiza (Sargassum), Zicai (Porphyra) used to for the treatment of scrofula, goiter, tumor, edema, accumulation, testicular pain and swelling. In addition, crude extracts of *Ulva lactuca*, *Monostroma greggii*, *Enteromorpha intestinalis* and *Palmaria palmate* can be used as cold compresses for nosebleeds, migraines, burns, sores and cuts. Laminarina obtained from *Laminaria angustata* (brown algae) used as hypotensive agent. Chloroform from *Chlorella*, Vaca and Walsh from *Ascophyllum nodosum* used as antibiotics. *Codium tomentosum*, *Codium fragile*, *Jania rubens*, *Dictyota*, *dictyopteris* and some other algae contains different ingredients which are used as pharmaceutical cosmetics. Various cosmetic products such as creams and lotions are based on alginate or carrageenan which improves the skin moisture retention properties of the product [1, 16, 21-23].

Seaweeds are widely used as body wraps to clean the toxins, soften the skin, relieve stress and improve skin condition. The electrolytic magnetic action of the seaweeds releases excess body fluids from congested cells and dissolves fatty wastes through the skin replacing them with depleted minerals particularly potassium and iodine. Seaweeds bath are nature’s perfect body balancer and they are an excellent way to take in iodine therapy. Seaweeds purify and balance the ocean; they can do the same for our body. A seaweed bath can maintain the balance of hormone in a more youthful body. Seaweeds are high in fiber and water contents and thus they are good to reduce the risk of heart disease and control hunger. Other nutrients in seaweeds also improve optimal immune function, neurological function, thyroid function, bone health and muscular function. Seaweeds can help to prevent and treat cold, flu, muscular cramp and arthritis. Brown seaweeds may be effective in treating hyperlipidemia, herpes, hypertension and other infections.

Food

Different seaweeds like *Ulva lactuca*, *Wakame*, *Laminaria digitata* (Kombu/Kelp), *Himanthalia elongata*, *Palmaria palmate*, *Enteromorpha sp.*, *Dictyota sp.*, *Sargassum sp.*, *Turbinaria sp.*, *Caulerpa racemosa*, *Porphyra*, *Rhodymenia palmate* etc. are extensively used as food. Chinese, Japanese, Koreans and Indonesians use different types of seaweeds as their traditional food. In Indonesia, some brown algae like Dicryta, *Sargassum* and *Turbinaria* are also commonly cooked with coconut milk. In Thailand, *Caulerpa racemosa* and *Gracilaria crassa* are commonly used to prepare spicy sauces. Indian used *Porphyra* to supply the salt needed for their body. In Scotland, *Rhodymenia palmate* is known as dulse, in Ireland as dillisk and in Iceland as sol has been widely used for food for 12 centuries. Young stipes of *Laminaria* and *Alaria* have also been eaten in Europe and North America. In Philippines *Sargassum siliculosum* is used as vegetable [1, 16, 21-23]. Fucoidan is a sulfated polysaccharide found mainly in various species of brown seaweed such as *Cystoseira species*, *Kombu*, *Limu moui*, Bladderwrack, *Wakame*, *Mozuku* and *Hijiki* which are marketed as a nutraceutical and food supplement. Collagen is a protein obtained from plants, animals and seaweeds (*Codium tomentosum*, *Codium fragile* and other seaweeds). The gelatin used in food and industry is derived from the partial hydrolysis of collagen. In the West Indies and South Africa jellies are prepared from species of *Gracilaria* [1, 16, 21-23].

As Animal Fodder

Daily use of a small amount of seaweeds as animal fodder improves the animal’s ability to digest the food and strengthen the natural immune system. Seaweed is a universal type of food and all kinds of animals, birds, chickens, pets and fish can use it as a food. China, Japan and Korea are the major consumers of different seaweeds like *Laminaria*, *wakame*, *Undaria pinnatifida*, Nori, *Hizikia* and *Porphyra*. Different seaweeds like *Sargassum*, *Ascophyllum*, *Laminaria*, *Macrocytis* and *Phaeophytes* are used as animal fodder. They are used as a supplement of mineral, vitamin, carbohydrates, proteins and fats. In Hong Kong,
species of Sargassum are dried and used as pigfeed. In Europe, seaweeds are collected, dried and ground into meals and fed directly to animals or used as supplement to supply minerals. Eggs of hens fed on seaweeds meal have increased iodine content. Increased butter-fat content of milk was reported from the cattle whose diet is supplemented with seaweeds meal. Most of thefish and marine animals feed on algae and therefore they are the part of the food chain [1, 16, 21-23].

**Agar**

Agar is obtained from different brown and red algae. Agar is a polysaccharide which solidifies almost anything that is liquid. Agar was first used in China in the 17th century and today it is produced in Japan, Korea, Australia, New Zealand and Morocco etc. Now the most important worldwide use of agar is as a gelatin-like medium for growing organisms in scientific and medical studies. It is used in antacid to neutralize stomach acid. It is used as a stabilizer or emulsifier in a variety of products. In addition, agar is used in the finishing of paper which improve ink acceptance by paper. It gives the cream coloring into ice cream and makes it creamy. It mixes fat evenly and used for the dressing of salad. It also turns thin and runny liquids into thick and creamy. It allows lipstick to be luscious (easy to apply) and also used as moisturizers in skin cream. It refines the application of dye on textile [1, 16, 21-23].

**Alginate**

"Alginate or algin" is the term usually used for the salts of alginic acid, but it can also refer to all the derivatives of alginic acid and alginic acid itself. Alginate is present in the cell walls of brown algae as the calcium, magnesium and sodium salts of alginic acid. Alginate absorbs water quickly, which makes it useful as an additive in dehydrated products such as slimming aids, and in the manufacture of paper and textiles. It is also used for waterproofing and fireproofing fabrics, as a gelling agent, for thickening drinks, ice cream and cosmetics, and as a detoxifier that can absorb poisonous metals from the blood. Alginate is used in various pharmaceutical preparations such as Gaviscon, Bisodol, and Asilone. Alginate is used extensively as impression-making material in dentistry, prosthetics and lifecasting. It is also used in the food industry, for thickening soups and jellies. Calcium alginate is used in different types of medical products, including burn dressings that promote healing and can be removed with less pain than conventional dressings [1, 16, 21-23].

**Carrageenan and β-Carotene**

Carrageenan is a hydrocolloide which is obtained from red algae. It is a linear sulphated polysaccharide present in the cell-wall of brown algae which is used as a stabilizer or emulsifier and is commonly present in dairy and bakery products. It is used as infant formula i.e. it brews a better bottle for baby and thickens the ingredients. It is used as de-icer (wipes out windshield ice i.e. bonds and stabilizes ingredients for melting). It puts the paste in toothpaste and also absorbs water. It keeps pancake syrup the flow slow and steady. It is used in breads for better-browning, stay-moist and for fluffy loaves. It is used in air freshener gels to stabilize gels to release odors gradually. It keeps tomato sauces thick and saucy (solids don't settle to the bottom). Green algae are dominant in the β-carotene content. This natural pigment gives creamy and yellow appearance to the ice cream. It makes cheese look cheesy (yellow-orange) and it saves spread-ability and stops oil separation from peanut butter.

**Bio-fuels**

Algae can be used to produce bio-fuel with the higher yield. They can produce 30 to 100 times more energy per hectare compared to terrestrial crops. However, bio-fuel from algae is quite expensive. Algae are considered a promising potential feedstock for next-generation bio-fuels, as certain species of algae contain high amounts of oil. Different biofuels like biogas, bio-alcohol and biodiesel can be produce from both microalgae and macroalgae (Scheme-1). Production of oil from algae is the next biggest opportunity for the bio-fuel industry. Algae like other plants use photosynthesis to convert solar energy into chemical energy. They store this energy in the form of oils, carbohydrates and proteins. Algae derived bio-ethanol is the alternative of gasoline to power the vehicles. This biomass source has a potential to produce the biogas on large scale that can be further converted to electrical energy. Algae are the rich source of bio-hydrogen which is one of the renewable and green fuels. Production of bio-fuels from marine algae is commercially more important but it is less explored.
area which demands the extensive research to make this technology viable [1, 16].

**Diatomaceous Earth**

The typical chemical composition of diatomaceous earth is 86% silica, 5% sodium, 3% magnesium and 2% iron. This product comes from the large fossil deposits of planktonic algae called diatoms. One of the largest sites of diatomaceous earth is in Lompoc, California. This material is actually the silica cell walls of these protists (walls that have minute pores). It is used as a filtration aid, as a mild abrasive, as a mechanical insecticide, as an absorbent for liquids, as an activator in blood clotting studies and as a component of dynamite. As it is also heat-resistant, it can be used as a thermal insulator. Products containing diatomaceous earth are sensodyne, tooth paste (for sensitive gums), silver polish and swimming pool filter powder.

**Fertilizers**

Algæ are the important source of potassium compounds therefore, a variety of raw seaweeds have been used as fertilizers in many countries like Japan, China, UK, Canada and Norway etc. Some algae like *Gracilaria verrucosa* and *Laminaria* species are burned to obtain soda and potash. As a fertilizer, potash was used for the growth of soybeans, tobacco, potatoes, sugar beets and corn. The beneficial effects of seaweeds are probably due to the alginate in brown seaweeds which increase soil structure and water holding capacity of the soil. The high potassium content of brown seaweeds like *Sargassum*, *Turbinaria*, *Hormophysa* and *Hydroclathrus* make them ideal substitutes for costly fertilizer. Several scientific studies have proved the effectiveness of liquid seaweeds extracts, which are now widely accepted in the horticultural industry. When applied to fruit, vegetable and flower crops, improvements have included higher yields, increased uptake of soil nutrients, increased resistance to certain pests such as red spider mite and aphids, improved seed germination and greater resistance to frost. Chitosan is mostly obtained from brown algae, shellfish, crabs, lobster and shrimp used as a natural seed treatment and plant growth enhancer, and as ecologically friendly bio-pesticide substance that boosts the innate ability of plants to defend themselves against fungal.
infections. The crude extract of Stonochloropera
marginatum is reported to kill the root-knot nematode
named as Meloidogyne javanica [1, 16, 21-23].

As Shelter

They also provide shelter and a home for
numerous fishes, invertebrates, birds and mammals.
Large seaweeds can form dense underwater forests,
called kelp forests. These forests provide a physical
structure that supports marine communities by
providing animals with food and shelter. Kelp forests
act as underwater nurseries for many marine animals,
such as fish and snails. The lush blades form a dense
forest canopy where invertebrates, fishes, birds, otters
and whales can find lots of tasty food and a good
home. Beautiful sea slugs and kelp crabs can be seen
on the blades and stipes of the seaweeds, while other
small marine animals like worms find their homes in
the holdfasts. Kelp forests are a huge food source
for sea urchins and other grazing invertebrates [18-20].

Terpenoids

Terpenoids are secondary metabolites
synthesized by plants, marine organisms and fungi by
head to tail joining of isoprene units [24]. They are
also found to occur in rocks, fossils and animals
kingdom. Terpenoids find wide applications in
industry such as linalool along with the phenyl-ethyl
alcohol is used in perfumery [24]. Citral is used as
mosquito repellent and the starting material for the
synthesis of vitamin A, menthol and artemisinin in
pharmaceutical industries [24]. Farnesol and
dalvionene are insect juvenile hormones whereas
cydosene are insect anti-moulting hormones [24].
Gibberellic acids and brassinolides are the plants
growth stimulants. Salamin and azadirachtins are
insect antifeedant and growth inhibitors. Taxol and
cucurbitacins are anti-tumor compounds [24].
Forskolin is a unique adenylate cyclase stimulator
which displays a variety of biological activities
including blood pressure lowering, positive inotropic,
hypolipidemic, antiglaucoma etc [24]. Artemisinin
is a sesquiterpene peroxide with potent anti-malarial
activity. Finally, derivatives of panaxadiol and
panaxatriol are immunostimulants. Natural rubber is
a well known commercial product is also a polymer
of isoprene unit. Moreover, insects use many
terpenoid-derived molecules for their communica-
tions [24].

A number of terpenoids (1-32) have been
isolated from various marine algae collected from
the coastline of Pakistan. The marine algae which have
been investigated for the isolation of terpenoids
include Laurencia pinnatifida, Jolyina laminariaeides,
Stokeyia indica, Codium intergarnii, Codium
decorticatum, Veloniopsis pachymena, Bryopsis
pennata, Cauierpa taxifolia and Dictyota dichotoma.
The phytochemical investigation on these algae has
been resulted in the isolation of 32 terpenoids mostly
belonging to the class of sesquiterpenoids and
diterpenoids. The marine red alga Laurencia
pinnatifida belongs to the genus Laurencia and
family Rhodomelaceae. It founds in the intertidal
zones of Karachi coast, usually as a small and
isolated organism. A number of interesting
sesquiterpenoids such as pinnatane (1), pacifinol
(2), 6-ethyl-10-dibromo-3-chloro-7β,8β-
epoxo-α-chamigren (4), 4,10-dibromo-3-chloro-
7β,8β-epoxy-α-chamigrene (5), pinnatifidone (6),
1β,4β,10β-tri-bromo-3-chloro-7(14)-ene-α-chamigrene
(7), laurocit (8), pinnatinone (9), pinnatiferol (10)
and pinnatifigene (11) have been isolated from
Laurencia pinnatifida. In addition, a diterpenoid,
isophytol (12) was also isolated from this alga [2,25-
38]. The investigation on brown algae, Dictyota
dichotoma, has led to the identification of fourteen
diterpenoids namely, dictyol-A (13), dictyol-B (14),
dichothetraol (15), dichotopentaol (16),
[4R,8S,9R,14S]-4α,8β,9α,14α-tetrahydroxy dolast-
1(15)-ene (17), dichotone (18), dichotolen-A (19),
dichotolen-B (20), dichotolen-C (21), dichotodione
(22), his clandestine acetate (23), dichotolenone A (24),
dichotolenone-B (25) and one monoterpenoid namely,
doliscopal (26) [2,39-44]. Compounds 18 and 20
exhibited significant biological activities in anti-
bacterial, anti-fungal, Brine Shrimp and Lemna minor
bioassays [43]. Codium decorticatum (Woodw.) is a
green alga belongs to the family Bryopsidophyceae
was collected from Karachi coast of Pakistan. Four
acyclic diterpenoids such as 7,11,15-trimethyl-3-
methylene-hexadecan-1,2-diol (27), 3,7,11,15-
tetramethyl-hexadec-2-en-1-ol or trans-phytol (28),
3,7,11,15-tetramethylhexadecan-1,2-diol (29),
3,7,11,15-tetramethylhexadec-3-en-1,2-diol (30) and
3,7,11,15-tetramethylhexadec-1-en-3-ol or isophytol
(12) have been isolated from this alga. All these
compounds are related to the class of phytol [45].
Valoniopsis pachymena (Mart.) B8orn is a marine
alga that has been resulted in the isolation of two
compounds namely, [2, 3-epoxide-3,7,11,15-
tetramethylhexadecan-1-ol (31) and \textit{trans}-phytol (28) [45]. \textit{Bryopsis pennata} Lamour belongs to the family Bryopsidophyceae. Chemical investigation on this alga has been resulted in the identification of two acyclic diterpenoids namely: 7,11,15-trimethyl-3-methylene-hexadecan-1,2-diol (27) and \textit{trans}-phytol (28) [45]. Chemical investigation on \textit{Canterpa taxifolia} has resulted in the identification of 7,11,15-trimethyl-3-methylene-hexadecan-1,2-diol (27) and \textit{trans}-phytol (28) [45]. \textit{Codium lyngari} is a green alga belongs to the genus \textit{Chlorophylla} is rarely studied and found complex with reference to their chemical composition which might be due to the presence of much chlorophyll. Two diterpenoids namely, \textit{trans}-phytol (28) and codioside ester (32) were isolated from this green alga [46]. \textit{Jolyja laminarioides} Guimaraes [syn. \textit{Edarachne binghamiae}] is a dark brown alga collected from the coastline of Karachi has been resulted in the identification of \textit{trans}-phytol (28) [47]. \textit{Stokeyta indica} Thivy et Doshi is a benthic brown alga occurring in mid to lower littoral pools with rocky bottom along the Karachi coast of the Arabian sea. It belongs to the genus \textit{Cystosperma} which like other brown algae has been the target of extensive phytochemical investigations. Chemical investigation on this alga has been resulted in the isolation of a monoterpenoid, \textit{loliolide} (26) [48].

\textbf{Steroids}

Steroids are compounds possessing a characteristic tetracyclic carbon skeleton, named as perhydrocyclopenteno phenanthrene nucleus or sterane [49]. They include a wide range of naturally occurring compounds like the sterol, bile acids, sex
hormones, adrenocortical hormones, cardiac glycoside, sapogenins and some alkaloids [49]. These steroids have the number of function in human physiology and are of immense biological importance. The most important compound in this class is cholesterol [49]. It is the most abundant steroid present in humans and most important one because all other steroids are derived from it [49].

A number of steroids (33-50) have been isolated from various marine algae collected from the coastline of Pakistan. The marine algae which have been investigated for the isolation of steroids include Jolyna laminarioides, Spataglossum variabile, Stokkeya indica, Dictyota indica, Laurencia obtuse, Melanothamus aflaghussaini, Codium iyengarii and Iyengaria stellata. The phytochemical investigation on these algae has been resulted in the isolation of eighteen steroids. Jolyna laminarioides Guimaraes [syn. Edarachne binghamiae] is a dark brown alga has been resulted in the identification of four steroids namely, fucosterol (33), 24-methylenecholesterol (34), sarangosterol (35) and 24-methylenecholesta-5,25-dien-3β-ol (36) [1-3, 47, 50, 51]. Compound 33 exhibited activity against Curvularia lunata, Stachybotrys atra and Microsporum canis [37]. Spataglossum variabile Figari et De Notar is a brown alga belongs to the family Dictyotaceae and grows on mid and littoral rocks along the coastline of Pakistan near Karachi city. Phytochemical investigation has led to the isolation of number of structurally interesting compounds, namely varninasterol [37], spatosterol (38), fucosterol (33) and cholesta-5-en-23-yn-3β-ol (39) [2, 3, 52, 53]. Stokkeya indica Thivvy et Doshi is a brown benthic alga has been resulted in the isolation of two compounds namely, fucosterol (33) and β-sitosterol (40) [2, 3, 54]. Dictyota indica is a brown alga belongs to the genus Dictyota which extensively found along the coast of Karachi city. A steroidal compound sargasterol (41) has been isolated from this alga [2, 50]. Laurencia obtuse (Hudson) Lamouroux is a marine red alga belongs to the family Rhodomelaceae has been collected from the coast of Pakistan has resulted in the isolation of cholesterol (42) as a major metabolite [2,50]. Melanothamus aflaghussaini [syn. M. somalisens] is a reddish brown marine alga which belongs to the order Ceramiales. A number of interesting compounds such as somalenone (43), cholesterol (42), 7-oxo-cholesterol (44) and 24-methylenecholesterol (34) were isolated for the first time from M. aflaghussaini [2, 51]. Codium iyengarii is a member of Chlorophyta which has been resulted in the identification of different compounds such as clerosterol (45), iyengadione (46), iyengaroside-A (47), iyengaroside-B (48) and clerosterol galactoside (49) [2, 50, 55]. Compounds 46-50 were screened for anti-bacterial activity but only compounds 47 and 49 were reported to be active against some bacteria [55]. Iyengaria stellata Børgeesen is a brown alga belongs to the order Scytosiphonales. It commonly grows on the rocks in the upper and mid-littoral regions along the coast of the Northern Arabian sea. The methanolic extract of this alga showed weak anti-bacterial activity against Streptococcus pyogenes at a concentration of 100 μg/100 ml and anti-fungal activity against Microsporum canis, Pleurotus ostreatus, Trichophyton rubrum, Drechslera rostrata, Nigrospora oryzae and Trichophyton simii at concentrations of 200μg/mL. A steroidal compound saringosterol (50) has been isolated for the first time from I. stellata [31].

Conclusions

Through the extensive literature of the marine algae with respect to their classification, ecology, biodiversity and applications it was concluded that the marine algae has a vital role for the sustainable life around the globe. The major uses of algae as medicines, biofuels, food and fertilizes can undoubtedly change the present crisis in these areas. The literature survey showed that different algae contain different structural types of terpenoids. It has been observed that sesquiterpenoids and diterpenoids are predominant over other classes of terpenoids. The skeleton of terpenoids isolated from Laurencia pinmitifida is entirely different to that of Dictyota dichotoma which in turn different to that of phytools isolated from Codium decorticatum, Veloniopsis pachynema, Bryopsis pinnata, Caulerpa taxifolia and Codium iyengarii. Some of the algae are rich in terpenoids and some in steroids such as Laurencia pinmitifida, Dictyota dichotoma and Codium decorticatum are rich in terpenoids whereas Jolyna laminarioides, Spataglossum variabile and some other algae are rich in steroids. In general, natural occurrence of terpenoids was found to be more than that of steroids. It was concluded that some of the terpenoids and steroids exhibited biological activities to a little extent.

Acknowledgements

The author is greatly thankful to Prof. Dr. Muhammad Shaiq Ali, International Center for Chemical and Biological Sciences (ICCBS),
University of Karachi, Pakistan for the provision of literature and valuable suggestions. The author also acknowledged his Ph.D. students, Shamsa Naz and Sumaira Noreen for their support in the preparation of the manuscript.

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