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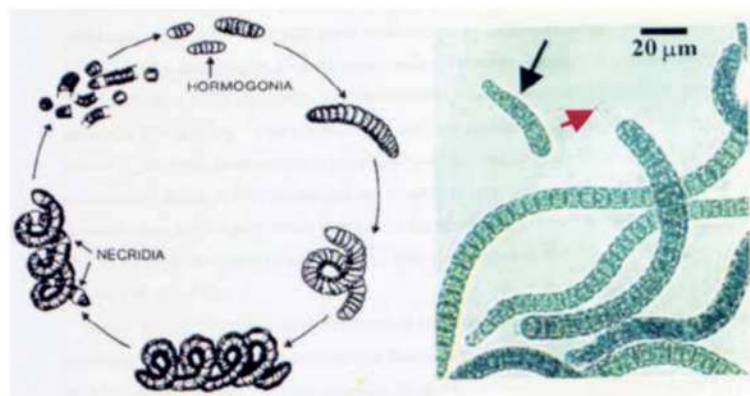
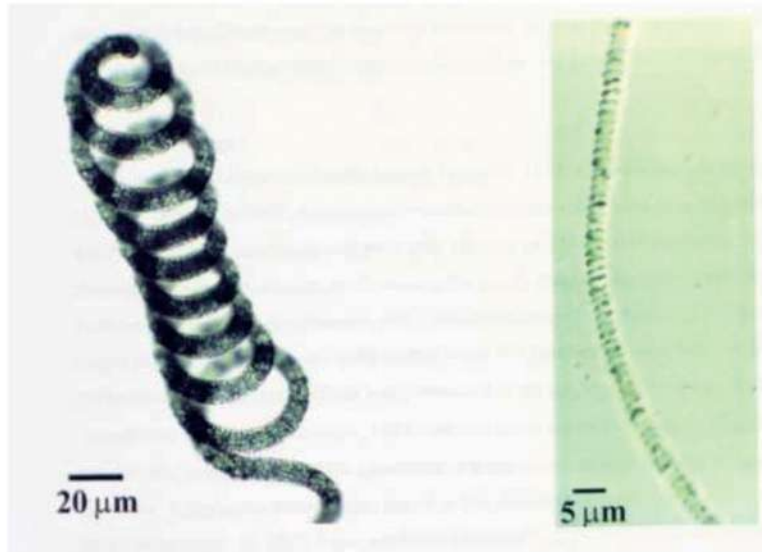
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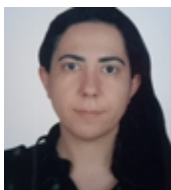
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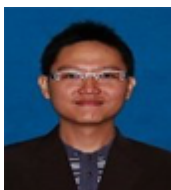
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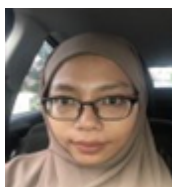
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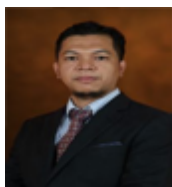
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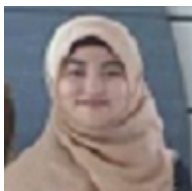
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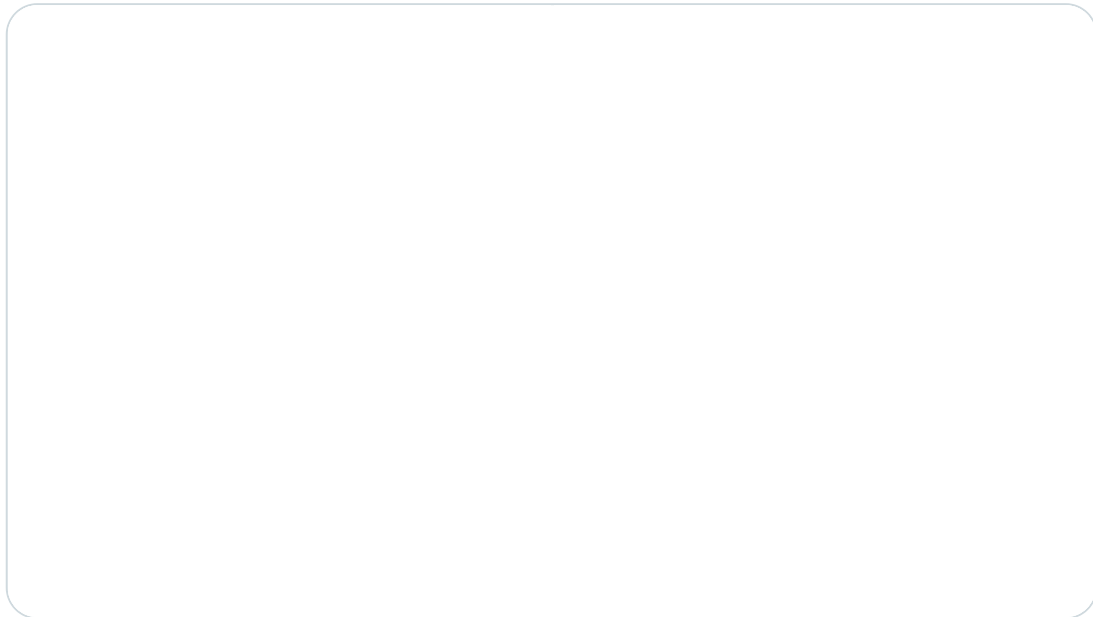
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
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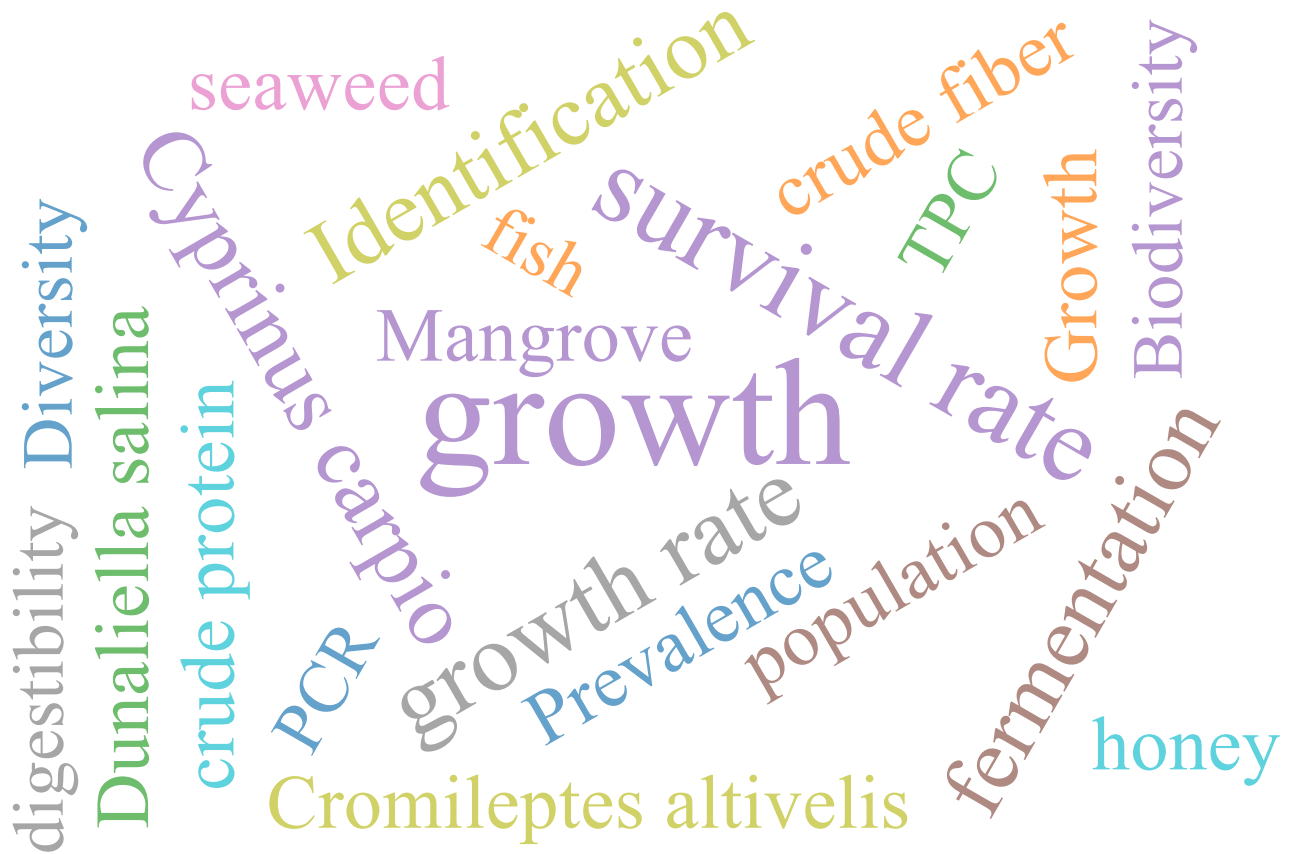
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Review Article

Trends in the Uses of Spirulina Microalga: A mini-review

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Abstract

There is a need to have a single document that summarises the present day uses of Spirulina. In this review, the research trend on the health and other applications of Spirulina microalga was critically evaluated. In terms of the health uses, antioxidant, antibacterial, and immunostimulant effects of Spirulina were emphasized. Other uses of the microalga discussed include the use of Spirulina for human and animal food, bioenergy, pollution and ecotoxicology control, cosmetics, bioplastics and biofertilizers. Literature search revealed that Spirulina polysaccharides, phycocyanin size and content play a role in antioxidant activity and DNA repair. The double bonds and positions of –COOH and –OH in Spirulina phenol content and γ -linolenic fatty acids (γ -LFA) have antimicrobial activity. Some compounds in Spirulina improve immune, increase survival rate and enhance distribution of proteins like hepcidin and TNF- α in animal models. High protein, vitamins, fatty acids (FAs) and glycoproteins in Spirulina are easily digestible due to its lack of cellulose and can improve human and livestock growth. Spirulina produces biodegradable and non-toxic biodiesel and useful co-products. Absorption of heavy metals by chemisorption occurs in Spirulina. Phycocyanin and β -carotene of Spirulina increase skin health, Spirulina also cause high cell proliferation and aids wound healing. Bioplastics produced from Spirulina are biodegradable, non-toxic with high blends. Biofertilizers from Spirulina have little or no residual risks, adds soil Nitrogen through Spirulina Nitrogen fixation ability. In addition, the survey of published works on Spirulina for the past two decades indicates that more research is been carried out in recent years using Spirulina, especially studies involving its health potentials and those concerned with molecular analysis. In conclusion, Spirulina is an exceptional commodity with numerous applications, and probably, some of its compounds causing those effects are yet to be isolated and that is one area for further research.

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1. Introduction

Microalgae, equally known as phytoplankton refer to all the aquatic autotrophs that live in suspension in the water column and the name encompasses several phyla which are mostly eukaryotes with the inclusion of photosynthetic prokaryotes called cyanobacteria (Widowati *et al.*, 2017). Microalgae are obviously very important for their role in food production in aquatic ecosystem as components of the food chain. Early studies on phytoplankton were focused on their use for food by fish and other aquatic organisms, both in their natural habitats and in aquaculture (Brown *et al.*, 1997) however, microalgae are being used for other numerous purposes presently. In mid 2000s, the world Spirulina production for human use was estimated to stand at more than one thousand metric tons annually (Khan *et al.*, 2005). However, it is obvious that more production is needed since it has numerous uses including the pharmaceutical industry where a major challenge with drug active ingredients from natural sources is their insufficient supply. In terms of quantity produced, USA is reported to be leading, then Thailand, India, Japan followed by China (Khan *et al.*, 2005).

Spirulina is a “blue-green” photolithoautotroph, meaning it is able to derive energy from sunlight and use carbon dioxide as its carbon source (Perry *et al.*, 2002). It obtains minerals from the inorganic sources of the environment and similar to other cyanobacteria, Spirulina is gram negative with a cell membrane, cell wall and an outer membrane. Spirulina has been known for its food benefits for numerous years even before its rediscovery three decades ago (Barnett, 2007) as the sale of little biscuits of dried Spirulina biomass known as “Dihe” decades ago in a market in present day Chad is a good example (Cliferri, 1983; Perry *et al.*, 2002). *Arthrospira maximus* obtained from Lake Texcoco (around present day Mexico) which was added into bread by the Spanish Conquistadors is another old instance of the use of Spirulina for food by man (Cliferri, 1983).

The phylum cyanobacteria otherwise known as “blue-green algae”, encompasses photosynthetic prokaryotes which have the capability to produce chlorophyll *a* (Whitton and Potts, 2000). The ability of cyanobacteria to photosynthesize “anoxygenically” in sulphide conditions has been established (Klatt *et al.*, 2015) however, on a general note; they use water as electron donor for photosynthetic process which results

in the release of oxygen. The taxonomic classification of cyanobacteria was done based on their ability to synthesize phycocyanin, a blue phycobilin pigment. The bluish colouration of these organisms which was the reason for their nomination as “blue-green algae” is as a result of the blue phycobilin pigment they possess (Müling, 2000).

The taxonomic relationship of cyanobacteria with bacteria has long been recognized since more than 100 years ago by researchers like Cohn as well as De Bary and also, between 1930 and 1965; the fundamental differences in organization of their cells and those of eukaryotes was elucidated by Stanier and Van Niel, as Stanier later provided a convincing submission that cyanobacteria possess the same prokaryotic cellular organization as do bacteria. His proof led to the current generally accepted nomination of cyanobacteria which can be traced back to 1979. In 1932, Geitler earlier produced a comprehensive treatise which recognized about 143 genera of cyanobacteria containing 1300 species on the basis of observations on field material, as reported by Müling, (2000).

The common species of Spirulina (*Arthrospira* and *Spirulina*) belong to the same order Oscillatoriales which encompasses all non-heterocystous, filamentous cyanobacteria that reproduce by binary fusion in plane without the production of alkinetes. The possession of *helical trichome* morphology by *Arthrospira* and *Spirulina* is what distinguishes them from oscillatoriacean genera. The life cycle of *Arthrospira* represents a typical and simple life cycle of the Oscillatoriales. The apex part of the trichome usually breaks as a result of the formation of specialized cells called *necridia*, the necridia are the ones that undergo cell lysis. The above account on *Arthrospira platensis* is presented as reported by Cifferi as well as Castenholz in the early and late 1980s respectively and as cited in Müling (2000). The necridia are distinguishable by their lack of pigments and bi-concave shape. Howbeit, irrespective of the strain concerned, necridia are not often noticed and fragmentation of the trichomes may hence occur without sacrificing a cell (Müling, 2000). The resulting cells called the hormogonia (usually short chains of cells or a single cell) are very motile, giving rise to a new trichome through the process of binary fusion which is often vertical to the longitudinal axis of the trichome.

A diagram showing the reproductive process in Spirulina as explained above is presented below in

Figure 2, before the diagram is the taxonomic classification of Spirulina (from kingdom to species) while the preceding diagram (Figure 1) shows some filamentous Spirulina cells under microscope (photo taken during one of our laboratory works).

Brief Biological Classification of *Spirulina platensis*:

- Kingdom : Eubacteria
- Subkingdom : Negibacteria
- Phylum : Cyanobacteria
- Class : Cyanophyceae
- Subclass : Oscillatoriothycidae
- Order : Spirulinales
- Family : Spirulinaceae
- Genus : Spirulina
- Species : *Spirulina platensis*

Source: Algae Base, last accessed: 17th April, 2019.

Raceways with shallow depths are commonly used for commercial production of Spirulina, howbeit; there are also scenarios of harvesting Spirulina from their natural populations for commercial use. “Semi-natural” cultivation involves harvesting during the day and night and allowing its biomass to double within 3-4 days. Filtration is done and the Spirulina biomass is homogenized and pasteurized followed by a spray-drying process (Oliguin, 1986).

Laboratory cultivation on other hand is mostly done in bottles at experimental scale as all the needed conditions for algal growth are provided artificially. Basically, 8 environmental factors influence Spirulina productivity including temperature of $\pm 30^{\circ}\text{C}$, stirring speed, luminosity, inoculation size, dissolved solids (10-60 g/litre), macro and micronutrients availability, pH of 8.5–10.5 and water quality (Ciferi, 1983; Ayala, 1998).

Unarguably, they are numerous research reports and reviews on the microalga Spirulina, however, single reviews discussing “all” its applications are still scarce. Also, newer applications of Spirulina have evolved over time; hence compiling a single review that discusses trends in its applications is very important. It is in this regard that this review was designed to discuss the present day uses of Spirulina from already published works

in reputable sources and from the practical experiences of the authors.

2. Use of Spirulina

Long before the development of current scientific knowledge, microalgae have played important roles in the life of human beings. Studies covering the trends in applications of Spirulina over different decades are presented below.

2.1 Health and Pharmaceutical Uses of Spirulina

Many works have revealed the health and pharmaceutical roles of Spirulina. Khan et al. (2005) reported that some species of Spirulina exhibit biomodulating and immunomodulating properties; *S. platensis* has a positive regulatory effect on immune system as it concerns the innate and specific immunity. Both human and animal immune systems have been reportedly enhanced via Spirulina intake (Khan et al., 2005) and many researchers have investigated and reported the anticancer and other effects of Spirulina (O’Shaughnessy et al., 2002; Grawish, 2008).

Spirulina as an Antioxidant Agent

The antioxidant ability of Spirulina and some other microalgae is well documented and this could be one of the reasons for their current popularity. According

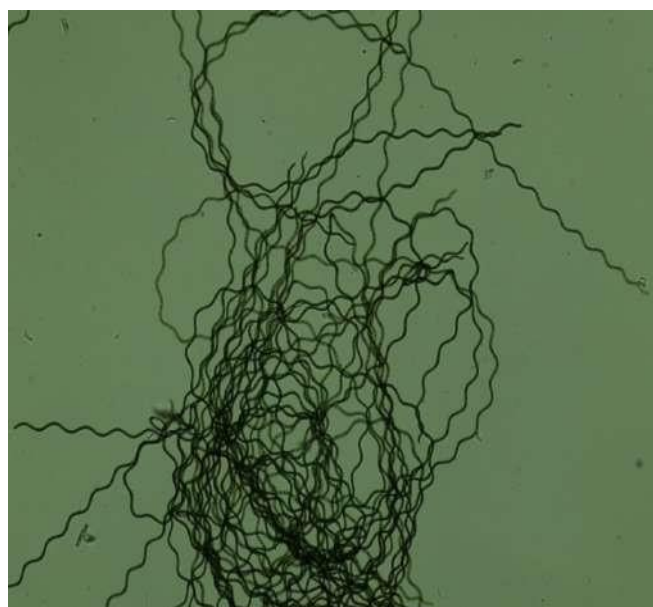


Figure 1. Microscopic view of *Spirulina platensis*. Authors’ Lab work (2019). Mag.=40x.

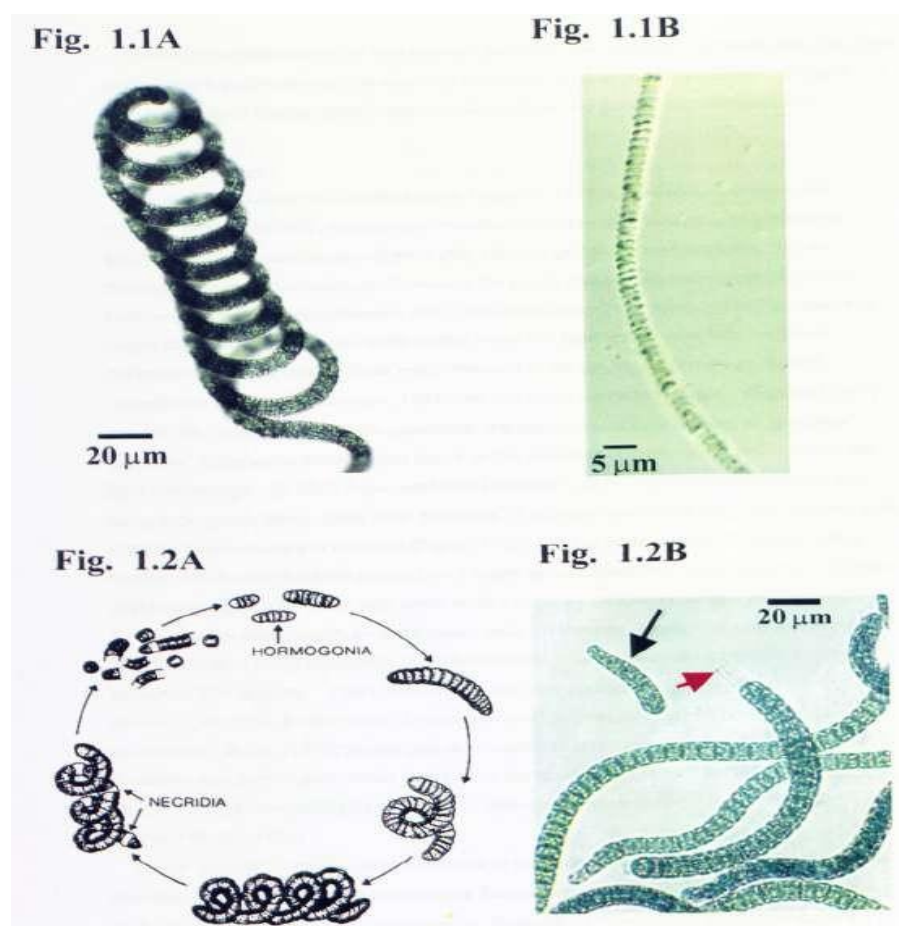


Figure 2. Microscopic image showing reproduction in Spirulina (*Arthrospira platensis*) (Müling, 2000).

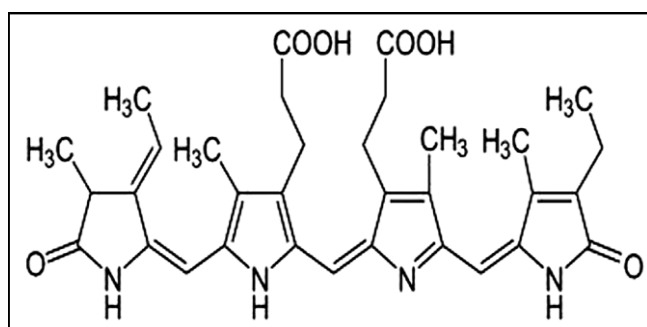


Figure 3. Chemical structure of C-phycoerythrin (Müling, 2000)

to O'Shaughnessy *et al.* (2002), carcinogenic processes can be reversed or interrupted with some specific agents prior to the onset of cancer; these agents could be synthetic or natural. The reports by Grawish (2008) revealed a tumor suppressive effect of Spirulina extract in hamster cheek pouch mucosa as a result of repair to the DNA damage.

The DNA repair is as a result of endonuclease activity that can be triggered by unique polysaccharide content of Spirulina (Grawish, 2008). Cyclooxygenase-2 (Cox-2) and Cyclooxygenase-1 (Cox-1) are the

two observed forms of a bifunctional enzyme (Hoseini *et al.*, 2013). Spirulina species produce C-phycoerythrin which functions as a selective inhibitor of Cox-2, the inhibition is known to occur as a result of the big and conformational structure of phycoerythrin, which aids its proper binding to the active spot of Cox-2 (Reddy *et al.*, 2000).

Recently, it has equally been revealed that selenium enriched *S. platensis* interrupted MCF-7 (human breast cancer cells) growth. The study by Bermejo *et al.* (1997) reported an excellent antioxidant activity in *S. platensis* as their work revealed protein extracts of *S. platensis* that scavenged peroxy and hydroxyl radicals and equally had an inhibitory activity on lipid peroxidation. Additionally, reports by Gad *et al.* (2011) indicated the strong inhibition of ferrozine-Fe²⁺ complex formation brought about by the chelating activity of Spirulina which results from its antioxidant compounds which are electron donors.

Spirulina as an Antibacterial

Since Spirulina contains many compounds including those with chromophore groups and bonds, this could form the bases for a thought about antimicrobial

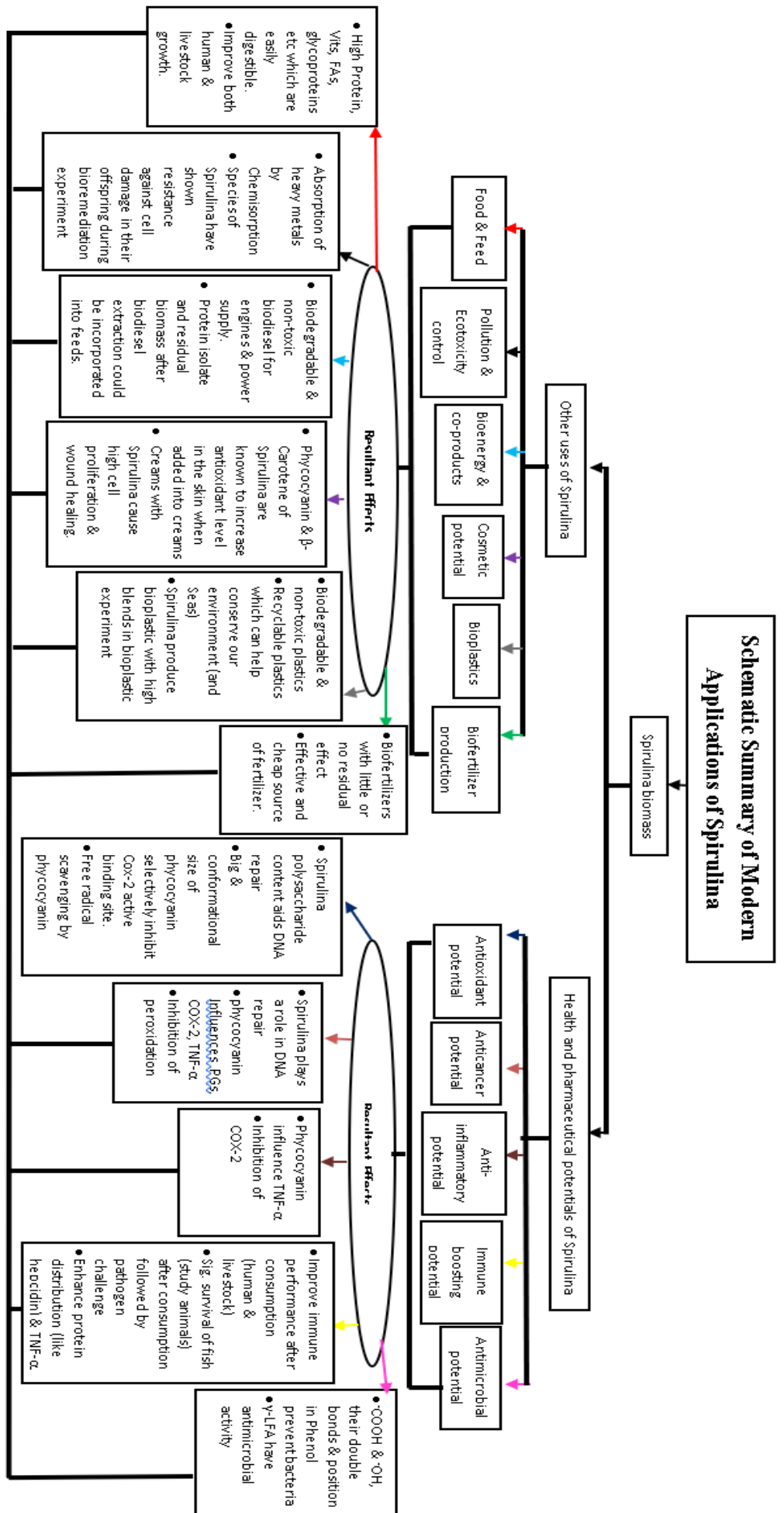


Figure 4. Conceptual Frame Work on Research Trends on the Uses of Spirulina

activity in Spirulina apart from other medicinal potentials often demonstrated by Spirulina. The study by [Demule et al. \(1996\)](#) observed that antimicrobial activity of methanolic extract of Spirulina is as a result of the availability of γ -linolenic fatty acid which is an active antimicrobial fatty acid available in high quantity in the alga extract.

The antimicrobial activities of Spirulina extract on some bacteria; *Escherichia coli*, *Staphylococcus aureus*, *Aspergillus niger* (a fungus) and *Candida albicans* (yeast) indicated that *C. albicans* were most sensitive to all the fractions of Spirulina tested ([Mendiola et al., 2007](#)). According to [Hoseini et al. \(2013\)](#), such antimicrobial activity may have a relationship with the synergistic effect of fatty acids. In addition, some studies on the antibacterial effect of Spirulina have proven it superior to some other natural antibacterial; [Ali & Doumandji \(2017\)](#) reported that methanol extract of Spirulina had a wider-spread spectrum of antimicrobial activities and appeared more promising against Gram positive bacteria compared to *Chlorella* which appeared more promising on Gram negative bacteria.

[Manigandan and Kolanjinathan \(2017\)](#) who studied the antibacterial activity of different solvent extracts of Spirulina using the disc diffusion and agar well diffusion methods reported higher antibacterial activity in methanol extracts using both methods however the results of [Nege et al. \(2020\)](#) on antibacterial effects two natural products showed that hexane fraction had the highest inhibition zone on *Staphylococcus aureus* among the tested crude and fractional extracts from Spirulina. These varied observations are perhaps pointers to the fact that different solvents and bacterial groups (Gram negative or positive), geographical location amongst other factors can influence the antibacterial activities of a natural product like Spirulina.

From our literature search, not many literatures focused on the antibacterial mechanism of Spirulina specifically, as most reports generally explained the antibacterial mechanism of microalgae. [Katircioglu et al. \(2005\)](#) stated that the antimicrobial activity of microalgae can be elucidated by the presence of cyclic peptides, lipopolysaccharides and alkaloids, the activity maybe as a result of toxins produced by microalgae cells since some blue green algae are known to produce toxins with potent pharmaceutical uses ([El-Sheekh et al., 2014](#)). [Alves et al. \(2013\)](#) submitted in their study on the mechanism of anti-MRSA that only phenolic acids (benzoic and cinnamic acid derivatives) were the main compounds with anti-MRSA activity at a point in their research, illustrating the relevance of the carboxylic group in the molecular structure (proton recipient). The report by the authors added that all the compounds

with anti-MRSA activity possessed an -OH (proton donor) and an -OCH₃ (proton acceptor) groups in the *para* and *meta* positions of the benzene ring respectively and the activity was observed to decrease in the absence of -OCH₃ group in the meta position (*p*-coumaric acid). However, the absence the above group in the structure of 2,4-dihydroxybenzoic acid was seemingly overlapped by -OH substitution in *ortho* position of the benzene ring. In addition, only -OCH₃ (proton acceptor) or H in position 5 of the benzene ring permitted anti-MRSA activity since the activity disappeared upon presentation of -OH in that position.

Although the mechanism explained by [Alves et al. \(2013\)](#) was using phenolic compounds identified in wild mushrooms, there is a tendency that the antibacterial mechanism of phenolic compounds from Spirulina will follow a similar pattern.

Immunostimulant and Growth Promotion in Animals

The relatively cheap cost of Spirulina makes it a good additive in animal feed for improved growth and immunity amongst other reasons. The study by [Abu-Elala et al. \(2016\)](#) and those of others revealed improved immune system in fish fed Spirulina supplemented diet even after a pathogen challenge compared with groups fed "normal" diet. [Abu-Elala et al. \(2016\)](#) recorded a significant survival rate and the distribution of proteins such as TNF- α in Spirulina fed group against the control, these suggest that one or more of the components in Spirulina may have caused those positive effects. In addition, [Bashandy et al. \(2016\)](#) also demonstrated the ability of *S. platensis* dose of 300 mg/kg to reduce the oxidative stress, sperm abnormalities and testicular damage in rats induced by Arsenic.

The findings of the above authors justify the submission that Spirulina also has immunostimulant as well as growth promotion potentials. Similarly, [Amer \(2016\)](#) found that *S. platensis* supplemented groups revealed higher levels of Lysozyme, Catalase and IgG compared to others. Obviously the immunostimulant and growth mechanism of Spirulina in the systems of animal models seems to involve different and many channels, for example the study by [Macias-Sancho et al. \(2014\)](#) reported that Spirulina stimulated the immune system of shrimp by direct reduction in apoptosis while [Noman \(2018\)](#) stated that Spirulina has potential to augment components of mucosal and systemic immune system via the activation of non-specific immune. The aqueous extract of Spirulina have reportedly influenced defence (immune) system through enhancement of the activity of phagocyte as well as stimulation of natural killer (NK) cells ([Ravi et al., 2010](#)).

The findings by [Nakono et al. \(2003\)](#) may have earlier justified the reasons for increased animal

immune upon consumption or treatment with *Spirulina* as the authors observed that the lack of cellulose in the cell structure of *Spirulina* makes it easily digestible therefore increasing fish appetite, improving their feed intake and digestibility which in turn enhances the overall health of the fish that subsequently leads to the fish' ability to defend infections via stress level reduction.

2.2 *Spirulina* for Human Food and Animal Feed

Spirulina contain high quantities of valuable proteins, vitamins, indisputable amino acids, beta carotene, essential fatty acids, mineral components, polysaccharides, sulpholipids and glycoproteins among other components (Glazer, 1988; Tandeau de Marsac and Cohen-Bazire, 1997; Fairchild and Glazer 1994) hence the addition of *Spirulina* to diets can provide a wide range of valuable nutrients (Khan et al., 2005). All edible species of *Spirulina* have certain peculiarities, they are regarded as a functional group which refers to a product acquired from a natural source that when consumed, it is very likely to provide health benefit and enhance system functions. *Spirulina* also has a high content of B vitamins, minerals, calcium, magnesium, iron, potassium, manganese, and zinc.

Moreover, the study by Agustini et al. (2016) on addition of *Spirulina* on ice and soft cheese and their sensory analysis proved that incorporation at 1 and 1.2% *Spirulina* was best for cheese and ice cream respectively. The same author in 2017 (Agustini et al. 2017) found in another study that incorporation of 9% *Spirulina* revealed a significant effect in elasticity, fat, water, β -carotene, protein, ash, carbohydrate content and sensory (hedonic) effects.

Khan et al. (2005) referred to *Spirulina* as a suitable matrix for biotechnological incorporation of new food trace element preparation. It is an important source of essential fatty acid; the gamma linolenic acid (GLA) (Colla et al., 2004; Otles and Pire 2001) as 10 g of *Spirulina* harbours over 100 mg of GLA (Nichols & Wood, 1968; Roughhan, 1989). The high quantity of riboflavin (Vitamin B₁₂) in *Spirulina* is another reason for its great nutritional value since this can be a very useful source of vitamin B₁₂ to vegetarians who often lack this vitamin in their diets (Dagnelie et al. 1991).

It is not surprising that the United Nations (UN) referred to *Spirulina* as "the most ideal food for mankind", the San Francisco Medical Research Foundation called it "the immune system miracle worker" while the Food and Agriculture Organization (FAO) of the UN called it "food for the future" (Agrotech, 2012).

Applications of *Spirulina* in Aquaculture

As of date, numerous works have been docu-

mented on the use of *Spirulina* as a complete or partial supplementation for protein and/or other nutrients in aquaculture via aquaculture feeds. As far back as 1994, the reports of El-Sayed indicated that Seabream (*Rhabdosargus sarba*) fed on up to 50 % of *Spirulina* was indifferent and their feed conversion efficiency not superior to those fed exclusively fishmeal.

In 1996, Abalone (*Haliotis midae*) revealed significantly higher growth after being fed with *Spirulina* and fishmeal compared to feed made from soybean meal, casein, dried *Ecklonia maxima* and torula (Britz, 1996). Moreover, more recent works have equally supported the above observations on the incorporation of *Spirulina* into fish diets. El-Sheekh et al. (2014) who studied the effect of *Arthrospira platensis* on the growth and carcass of hybrid red Tilapia observed and recommended that 75% *Spirulina* can conveniently substitute fishmeal-based diet with increased feed conversion ratio as well as protein value in proximate composition of the carcass. A 10% replacement of fishmeal with *Spirulina* in the feeding trial of common carp (*Cyprinus carpio*) gave significantly higher body weight as against other treatments and the control (Abdulrahman et al., 2014), on the other hand Ibrahem et al. (2013) observed improved feed conversion ratio and growth rates in striped jack and Tilapia (*Pseudocaranx dentex* and *Oreochromis niloticus*). The findings of Amer (2016) showed that Tilapia supplemented with 1% *S. platensis* gave lower FCR but higher body weight compared to other groups and the control.

In addition, the study by Teimouri et al. (2013) on the effects of 0, 2.5, 5, 7.5 and 10% *S. platensis* and synthetic astaxanthin (50 mg) diets on the fillet and skin pigment including growth performance of rainbow trout (*Onchorhynchus mykiss*) and found that incorporation of 10% *S. platensis* gave the highest carotenoid deposition in fillet and skin, implying *Spirulina* can be used as a substitute for synthetic astaxanthin in the diets of rainbow trout. In terms of cost reduction in fish feed, El-Sheekh et al. (2014) observed that the utilization of *A. platensis* meal (75% inclusion) in the feed of red Tilapia resulted in a decrease in feed expenditure (cost/kg feed) and an incidence cost was accompanied with increased profit index. In addition, Nakono et al. (2003) earlier reported that the absence of cellulose in the cellular structure of *Spirulina* makes it easily digestible thereby increasing the appetite of the feeding organism (fish in the case of their study), improving both feed intake and nutrient digestibility which in turn enhanced fish health as well as increased their capacity to defend infections via the reduced level of stress. As a result of these positive findings, many authors in this area of have recom-

mended Spirulina as a good and economical nutritional supplement in aquaculture.

2.3 Bioenergy production

Indeed the rapid depletion effect of fuel obtained from fossils has given rise to the search for an alternative and sustainable fuel that can replace the “conventional fuel” to eliminate energy crises with little or no environmental effects (Rahman *et al.*, 2017). To achieve the above, many scientists are working day and night in search of eco-friendly and sustainable energy sources, hence the development of a renewable source of fuel still remains a serious issue globally.

Recently, biodiesel is becoming increasingly acceptable as an alternative available for researchers to supplement conventional fuel. Incidentally, the characteristics of biodiesel are highly similar to those of diesel and hence can fit at all points with diesel and biodiesel can be used in existing engines without a modification.

As a good feedstock, microalga is one of the most promising alternatives of conventional feedstock. Generally, algae contain a high level of oil compared to other feedstock. The report of Demirbas (2009) indicates that algae yields over 200 times oil per acre when compared to the best-performing plant or vegetable oils. In addition, biodiesel from algae is biodegradable, nontoxic, and renewable with great potential as a green alternative fuel for CI engine. It equally has acceptable combustion and emission profile compared with petroleum fuel (Mata *et al.*, 2010).

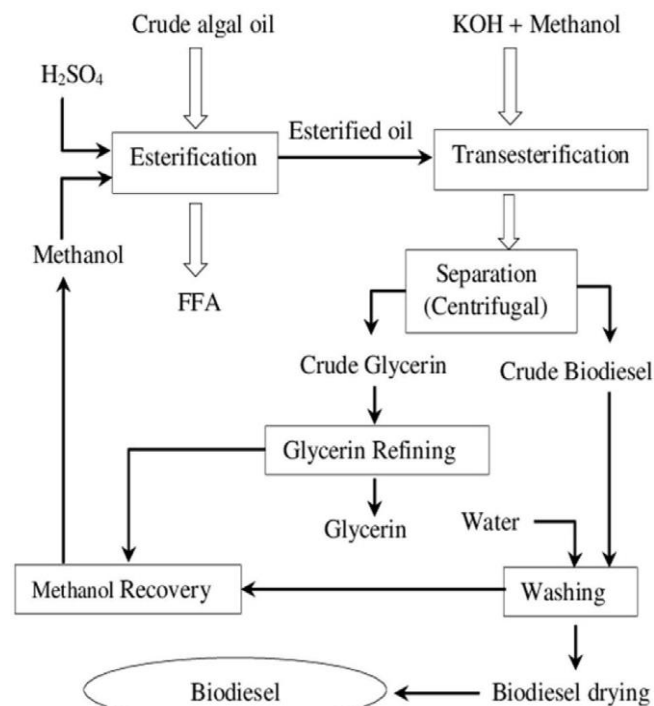


Figure 5. Biodiesel production flow chart (Rahman, *et al.*, 2017).

In the study on *Spirulina maxima* by Rahman *et al.* (2017) molar ratio which is one of the optimization parameters for esterification indicated that 3:1, 6:1, 9:1, 12:1 and 15:1 molar ratios at 65°C with 1.5% sulphuric acid at 350 rpm for a 2h reaction period gave good results of acid value (AV) reduction from 8.2 to 3 mgKOH/g with increased esterified oil yield to 22.2% during an increase in molar ratio from 3:1 to 12:1. However, further increase in molar ratio gave no reduction in AV, possibly due to water production effects (Berchmans and Hirata, 2008). Deng *et al.* (2010) also obtained the above 22.2% at 12:1 molar ratio in acid esterification of *Jatropha curcas* L. seed oil. According to Rahman *et al.* (2017) the optimum methanol to algal oil ratio was 9:1 since it reduced AV to the lowest; from 10.45 to 1.21 mg KOH/g.

2.4 Pollution and Ecotoxicology Prevention/Control

According to Murali and Mehar (2014), environmental pollution is but a product of industrial revolution. Environmental pollution is known to affect quality of life as well as environmental ecosystem. For several decades now, numerous attempts have been made to reduce environmental pollution and amongst those attempts is bioremediation. Bioremediation often involves the use of microorganisms for reduction of toxicity from harmful waste and heavy metals. This offers a solution to the management of solids by waste detoxification, the cost effectiveness of this technique and the environmental impact it offers makes it appear more attractive (Murali and Mehar 2014).

For the above reasons, various cyanobacteria species including *Spirulina* have been studied to assess their toxicity reducing ability. The ability of *Spirulina* to take up Dichlorodiphenyltrichloroethane (DDT) at concentrations of 10 and 50ppm was tested and it was able to assimilate approximately 65% (6-6.5 ppm) DDT at 10ppm and then about 80% (40ppm) of DDT at 50ppm concentration. Although more damage was observed on the *Spirulina* cells after incubation at 50ppm DDT apart from the detected residue but at 10ppm, less than 1% was observed (trace quantity of DDT).

The study by Chojnacka *et al.* (2005) on the mechanism of *Spirulina* in biosorption of metals including Cu^{2+} , Cd^{2+} and Cr^{3+} indicated that chemisorptions instead of physical adsorption was the main mechanism for sorption of heavy metals by *Spirulina*. Maximum contribution of physical adsorption in the study was only 3.7% while functional groups in the cell surface participated in the binding of metal ions by a biosorbent through equilibrium reaction. The findings of Rangsayatorn *et al.* (2002) showed that the uptake of heavy metals by *Spirulina platensis* was independent of the solution's tem-

perature but dependent on its pH as the optimum pH of biosorption was reported as 7. Meanwhile, the maximum adsorption ability of *Spirulina* was recorded as 98.04 mg Cd per biomass and the rate of heavy metal uptake was fast, 78% of the adsorbed metal occurred in the first 5mins, indicating a high rate of heavy metal absorption by *Spirulina*. Not so many scientific articles have clearly explained the mechanism of pollutant uptake by *Spirulina*, however, Dwivedi (2012) summarized the mechanism of metal uptake by algae as a process occurring via adsorption as follows, firstly, a physical adsorption which involves a quick uptake of metal ions over the cell surface in few seconds or minutes followed by a slow transportation of the ions into the cytoplasm in a process referred to as chemisorption.

2.5 Cosmetics

The term cosmetics is quite broad and covers substances used for all forms of body painting like for religious or ornamental purposes (Novak, 2010). The Resolution RDC no. 79 of August 28, 2000 defines cosmetics as products made from natural and/or synthetic substances to be used externally on human body parts such as hair system, nails, skin, teeth, mucous membrane of oral cavity and genitalia with the

aim of perfuming, cleaning, altering their appearance, protecting them, correcting or keeping them in good condition.

Antioxidant defence of the skin is dependent on synergistic effects of different antioxidants like vitamins (Vit C, Vit E isoforms), nutrition and endogenous enzymes (superoxide dismutase: SOD and catalase, and GSH peroxidase: GPx). According to Gunes et al. (2017), *S. platensis* is a microalga with high phycocyanin and the pigment phycocyanin has been largely used as a natural blue colourant in cosmetics. The results of Gunes et al. (2017) on the effect of *Spirulina* extracts in cream on cytotoxicity and wound healing activities revealed high cell proliferation at concentrations of 0.1 and 0.05%. Meanwhile the wound healing test showed a significant improvement as the groups administered cream incorporated with *S. platensis* extract improved significantly ($p < 0.05$) compared to the control, the effect was highest at 1.125% *Spirulina* extract with more efficiency on the skin cells and wound closure.

2.6 Bioplastics

Plastics are obviously one among the most commonly used commodities around us. Consumers of

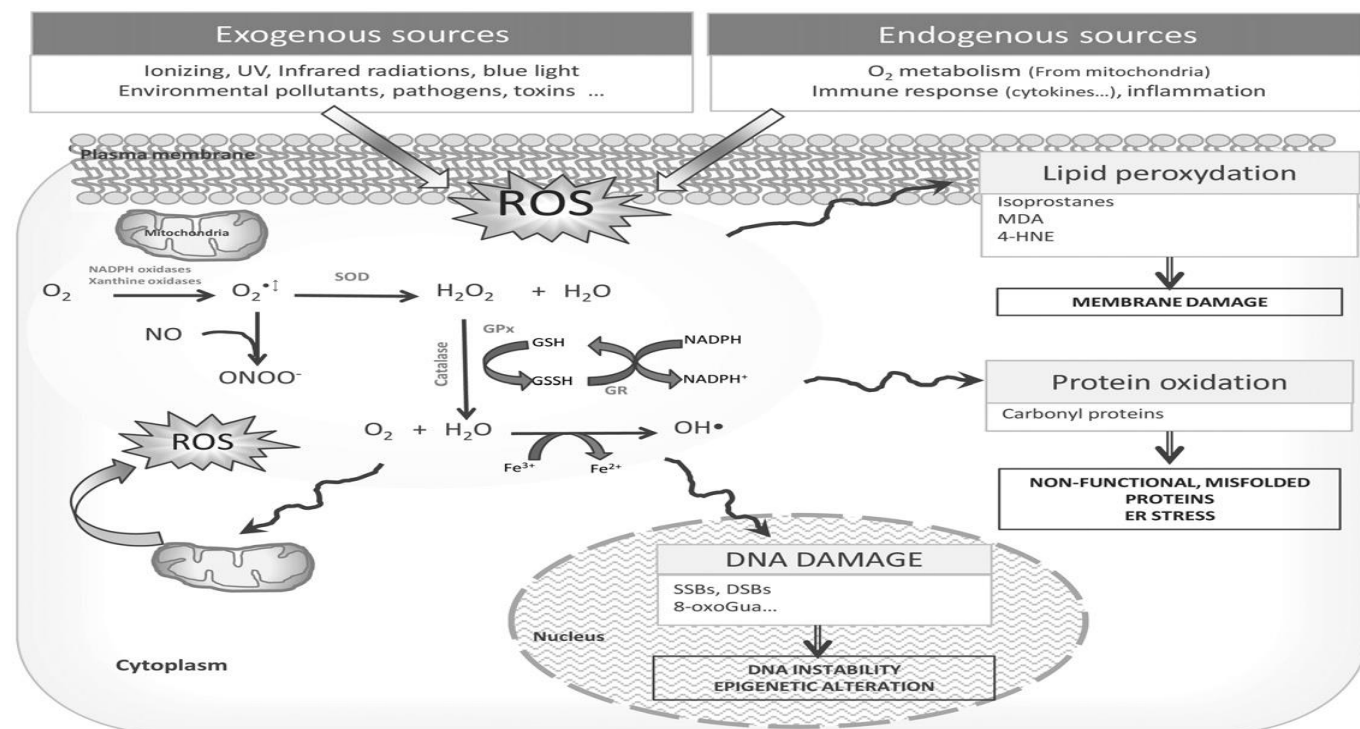


Figure 6. Summary of ROS pathways and their induced cellular damage. Cells of the skin face constant exposure to both endogenous and exogenous forms of ROS. Enzymes like xanthine oxidase or NADPH oxidase and the Mitochondria are endogenous sources of ROS. The cell is furnished with a variety of defense mechanisms for scavenging ROS. SOD catalyses the dismutation of superoxide resulting hydrogen peroxide while the reaction of catalase (CAT) with hydrogen peroxide trigger water and oxygen formation. Glutathione peroxidase (GPx) is known to reduce hydrogen peroxide. The reduced form of GSH is converted into oxidized glutathione (GSSG) at the point H_2O_2 is converted to water. Destroyed mitochondria yield more ROS in a reoccurring circle. Adapted from Berthon et al. (2017).

this age need inexpensive, versatile and convenient materials for the production of plastics (Zeller *et al.*, 2013). As at present, a major constituent of the world's plastic originates from petroleum which have a number of advantages such as high economies of scale and developed technologies (Iles and Martin, 2012).

Plastics are constantly used as a result of their strength which is combined with low weight, resistance to degradation by sunlight, water, bacteria and chemicals as well as their thermal and electrical insulation abilities (Zeller *et al.*, 2013). Polystyrene which is one of the most largely utilized plastics has a very slow degradation rate in our environment. High impact polystyrene (HIPS) as a copolymer of polystyrene and polybutadiene is a cheap, easy to manipulate and easy to fabricate type of plastic material.

Despite these “advantages”, the above plastic materials cause a great number of damages to the environment in different ways including waste production that lead to water, air and soil pollution. Some of these wastes are known to produce toxins capable of leaching over time, leading to contamination of ground water (Fraklin Associates, 2010). Biodegradable plastic raw materials have been obtained from foodstuffs like maize, rice wheat, potato and others but the land requirement and competition in food and feed needs is yet another challenge (Jerez *et al.*, 2007; Zeller *et al.*, 2013).

Microalgae like Spirulina on the other hand do not require large, fertile land and long period for growth. Spirulina houses numerous types of nutrients including a protein content of up to 60% on dry weight (Becker, 2007) which makes them a viable alternative to land crops. Also, their ability to pick up nutrients from wastewater and grow is another good advantage. Zeller *et al.* (2013) who studied bioplastic and thermoplastic blends from Spirulina and Chlorella microalgae revealed interesting results from their study. They developed blends of microalgal biomass and polyethylene from glycerol plasticization which was most effective at ratio 4:1 biomass to glycerol. In the above study, Spirulina blended better with polyethylene when compared to Chlorella samples; however, Chlorella was reported to exhibit higher bioplastic properties. The good performers of Spirulina in terms of blends led to their conclusion that Spirulina is more desirable for commercial applications.

In terms of tensile strength, the study by Dianursanti *et al.* (2018) revealed that increasing quantity of glycerol (plasticizer) decreased the tensile strength of bioplastics made from Spirulina, however, all the various percentages of glycerol addition tested gave higher tensile strength when compared with commercial plastic bags.

2.7 Biofertilizers

Probably due to population rise and technological advancement, fertilizers are heavily utilized in modern agriculture to increase output, however, the heavy use of inorganic and chemical fertilizers could form a threat to human health and the environment (Win *et al.*, 2018). Due to the aforementioned, efforts are being channelled into the exploitation of microorganisms as a more eco-friendly approach for conservational agriculture. Hence, biotechnological “tools” like microalgae, fungi, rhizobacteria which are able to live in beneficial association with higher plants are of great concern as potential sources of biofertilizer.

One good reason for using cyanobacteria as biofertilizers is their ability to fix nitrogen; these organisms are known to convert inorganic atmospheric nitrogen (N_2) into organic nitrogen which is easily utilized by higher plants (Kumar, 2016). Already, attempts have been made to use cyanobacteria in the promotion of the growth rate of rice in both Chile and India and the cyanobacteria indicated increased nitrogen accumulation efficiency in rice paddles (Singh *et al.*, 2016).

El-Rheem *et al.* (2015) who studied the stimulant effect of Spirulina under low level nitrogen fertilization on wheat plant observed that increasing levels of Spirulina microalga from 25–100 ml/L under low levels of nitrogen fertilization resulted in an increase in growth and yield of wheat without any negative effect from the decreased rate of nitrogen fertilization.

In addition to the above, the authors carried out an intense literature search on different applications of Spirulina globally in recent years reported by numerous reputable papers. The information is summarized in Table 1 below.

3. Conclusion

This review highlights the trend in Spirulina research and modern applications of Spirulina, and also, it has equally shown how important the microalga is to humans, livestock and the environment. Since these applications involve key areas of humanity and the environment, Spirulina appears a recommendable candidate, not just for medicines but also for food, biodiesel, cosmetics, bioplastics and biofertilizer production. In this age of environmental pollution and higher population, increased production of Spirulina and its conservative use will do humanity a lot of good. Efforts like Spirulina mass farming in landlocked countries and safe laboratory multiplication could increase its availability since some studies have already shown encouraging health potentials from microalgae clones. Isolation and studies on all yet to be known compounds in Spirulina are open areas for new research.

Table 1. Recent Findings on different Uses of Spirulina Documented in Different parts of the World over the last two Decades (2000-2020)

S/No	Research Aspect	Country/Region	Major Finding(s)	Reference
1.	Phytoremediation potential of Spirulina (biosorption and toxicity evaluation of Cadmium)	Thailand	Environmental factors were observed to have effect on biosorption. The optimum pH for biosorption of cadmium was 7. Rapid Cadmium uptake with 78% metal sorption in 5 minutes as the maximum Cadmium adsorption ability of Spirulina was 98.04mg Cadmium per biomass.	Rangsayatorn <i>et al.</i> (2002)
2.	Antioxidant properties under different temperature and nitrogen regimes	Brazil	Temperature of 35°C and sodium nitrate of 1.875 or 2.5g.L ⁻¹ had higher activity	Colla <i>et al.</i> (2007)
3.	Antioxidant activity and hepatoprotective effects of Whey and Spirulina <i>In vitro</i> and using rat model	Egypt	<i>In vitro</i> findings indicated a dose-dependent trend in antioxidant, metal-chelating and radical scavenging activities. Both natural products prevented liver damage by CCL ₄ but more pronounced effects recorded in rats that received a mixture of Spirulina and Whey.	Gad <i>et al.</i> (2011)
4.	Effect of Spirulina Bio-fertilizer on growth and yield of <i>Vigna radiata</i>	Myanmar	The highest leaf area index, leaf area ratio, crop growth rate, relative growth rate, maximum total dry matter, harvest index and yield were found in crops administered 7g/L of Spirulina suspension	Agung (2011)
5.	Bioplastics and their thermoplastic blends from Spirulina and Chlorella	Georgia	Spirulina indicated a good blend performance	Zeller <i>et al.</i> (2013)
6.	Bioactive substances from fresh and dried Spirulina	Indonesia	Higher flavonoid and phenolic contents in dried compared to fresh Spirulina. Lower IC ₅₀ for antioxidant test in dried compared to the fresh state.	Agustini <i>et al.</i> (2014)
7.	Antibacterial activity (<i>In Vitro</i>)	India	Water extracts of Spirulina had a higher activity than methanol, ethanol and acetone	Chakraborty <i>et al.</i> (2014)
8.	<i>In Vitro</i> Antibacterial activity	Bangladesh	Inhibition of bacterial growth	Ahsan <i>et al.</i> (2015)
9.	Antimicrobial (<i>In vitro</i>)	India	Effective against bacteria and fungi	Usharani <i>et al.</i> (2015)
10.	Antioxidant and anticancer activity	Egypt	Highest antioxidant activity and total phenolic content was recorded at highest concentration tested. IC ₅₀ of 18.8 and 22.3µg/mL was recorded for HCT116 and HEPG2 respectively.	Zaid <i>et al.</i> (2015)
11.	Effect of Spirulina on Wheat plants under low levels of Nitrogen	Egypt	Increase of Spirulina from 25 to 50 ml/L under low levels of nitrogen fertilization increased both the growth and yield of wheat.	El-Rheemkh <i>et al.</i> (2015)
12.	Optimization of protein extraction of Spirulina for generation of potential co-product and biofuel	Georgia	High protein yield (60%), Higher protein content in co-product (80.6%) while the residual biomass was lower in nitrogen content and higher in total non-protein content than the original biomass	Parimi <i>et al.</i> (2015)
13.	Effects of cosmetic containing Spirulina extract on young and mature skin	Brazil	Increased stratum corneum water content in adult and young groups given Spirulina cream. Significant reduction in the sebum content on volunteers' skin as well as an improvement in skin microrelief by reduction in surface roughness. More homogenous and uniformly distributed keratinocytes.	Delsin <i>et al.</i> (2015)
14.	Attached cultivation for improvement of Spirulina biomass productivity	China	High footprint areal biomass productivity of 60g/m ² /d was recorded. Similar nutritional content was found in the Spirulina cultivated with the new technique.	Lanlan <i>et al.</i> (2015)
15.	Microalgae cultivation for biosurfactant production	Brazil	Spirulina and other tested microalgae demonstrated potentials as organic natural sources of for triggering both microorganism growth and biosurfactant production.	Radmann <i>et al.</i> (2015)
16.	Mitigation of oxidative stress and repro-toxicity caused by Sodium arsenite in male rats	Saudi Arabia	Spirulina dose of 300mg/kg decreased induced oxidative stress, sperm abnormalities and testicular damages by its antioxidant potential.	Bashandy <i>et al.</i> (2016)

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|-----|--|------------|--|-----------------------------------|
| 17. | Evaluation of the wound healing and antioxidant activity of Spirulina extract added into skin cream (<i>In vitro</i> test) | Turkey | 0.1% of Spirulina extract gave higher proliferation activity compared to the control group with 198% cell viability after day 3. The skin cream with 1.125% crude extract of Spirulina demonstrated enhanced wound healing effect on HS2 keratinocyte cell line. | Gunes <i>et al.</i> (2016) |
| 18. | Application of Spirulina on ice cream and soft cheese and their sensory analysis | Indonesia | Addition of 1% and 1.2% Spirulina was observed as the best concentrations for incorporation into soft cheese and ice cream respectively. | Agustini <i>et al.</i> (2016) |
| 19. | Dietary supplementation with Spirulina and Garlic in Tilapia feed | Egypt | The mixture of Spirulina and Garlic significantly improved feed intake, live weight gain, specific growth rate, protein efficiency ratio and feed conversion ratio. The group also exhibited up-regulation of some immune related genes (TNF- α and liver hepcidin) and also recorded the least total mortality after bacteria challenge. | Abu-Elala <i>et al.</i> (2016) |
| 20. | Effect of the addition of various Spirulina concentrations into dried noodle | Indonesia | Dried noodles administered 9% Spirulina gave significant effect in elasticity; water, protein, β -carotene, fat, carbohydrate, ash content and sensory (hedonic) effects. | Agustini <i>et al.</i> (2017) |
| 21. | Effects of dietary supplementation of Spirulina and Chlorella on blood parameters of diabetic rats | Iran | Both Spirulina alone and its combination with Chlorella increased the levels of white blood cells (WBC), red blood cells (RBC), platelet, packed cell volume (PCV), selenium and glutathione peroxidase (GPx) in diabetic rats. | Emami & Olfati (2017) |
| 22. | Biodiesel production from Spirulina | Bangladesh | Optimum condition for maximum esterification yield was found to be at the molar ratio of 12:1 and temperature of 60°C. Biodiesel obtained were within standard limits when analysed with ASTM standards. | Rahman <i>et al.</i> (2017) |
| 23. | Effect of glycerol plasticizer in Spirulina based bioplastic | Indonesia | Optimum plasticizer composition for the Spirulina bioplastic was 30wt% with a tensile stress of 27.70kg/cm ² (close to commercial plastic bags) and 66% elongation. | Dianur-santi <i>et al.</i> (2018) |
| 24. | Spirulina as a tool against water pollution by 1,1'-(2,2,2-trichloroethane-1,1-diyl) bis(4-chlorobenzene); DDT | Georgia | 10ppm concentration of DDT is the acceptable for remediation by Spirulina in water. It was also observed that Spirulina removed about 70% of DDT after 15 days of incubation. | Kurashvili <i>et al.</i> (2018) |
| 25. | Spirulina in the treatment of fish farming wastewater | Brazil | Maximum cellular density of Spirulina resulted in the production of 0.22g/L of dry biomass with a maximum productivity of 0.03g/L. Ammonia, nitrate, nitrite and phosphate concentrations became lowered by more than 94.8%. | Nogueira <i>et al.</i> (2018) |
| 26. | Spirulina growth in photobioreactor under varied nitrogen concentration for maximised biomass, carotene and lipid contents | Egypt | Higher total carotene and total lipid contents were observed in nitrogen-limited condition | El Baky <i>et al.</i> (2019) |
| 27. | Nutritional analysis of Spirulina | Indonesia | Spirulina is a good nutritional source and a potential super food | Liestianty <i>et al.</i> (2019) |
| 28. | Effect of Spirulina on radiation-induced thyroid disorders and alteration of reproductive hormones | Egypt | Spirulina-treated group had a significantly attenuated oxidative stress in thyroid tissues, ameliorated DNA damage and decreased caspase-3 activity. | Ebrahim (2020) |
| 29. | Effects of Spirulina on the chemical, microbial and sensory properties of wheat flour pesta | Iran | Different levels of Spirulina inclusion had a significant effect on chemical parameters of pasta. Sensory evaluation showed that pasta incorporated with 0.25% Spirulina had a higher acceptability as against the control. | Mostolizadeh <i>et al.</i> (2020) |
| 30. | Antioxidative activity and phytochemical screening of <i>Spirulina platensis</i> , <i>moringa oleifera</i> and their synergies | Indonesia | Significantly higher antioxidant activity in the combined crude and hexane fractions from Spirulina and Moringa (synergism). Some phytochemical groups were also detected more in the mixture of the two. | Nege <i>et al.</i> (2020) |

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Authors' Contribution

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Conflict of Interest

All the authors of this review declare that they have no conflict of interest.

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