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Nutritional perspective of novel protein food sources a systematic review

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Abstract
Novel Protein Foods (NPFs) are products based on vegetable protein and micro-organisms.
The aspire of this paper is to explore and review the state of the nutritional profile of major novel protein sources for the future perspective as they become the alternative sources of animal protein particular spiralling, insects, rapeseed, duckweed, single cell protein, microalgae, and seaweed except that a wide variety of vegetarian alternatives is already available on the market: seitan, tofu, soy meat, tempeh, quoin and meatless based on lupines. As they are rich in protein content, high fibre, low in calorie, micronutrient supply of body can be easily met by the consumption of novel protein sources. The demand of novel food sources become increasing day by day because of International travel, Globalization, Changing Tastes, Increasing Desire to taste new food instead of traditional one, Advances in genetics traits, Innovation in field of plant breeding, Rapid increasing population, Increasing demands of food resources, Problem of under-nutrition. Novel protein food sources that can nourishes our body by providing most of the protein you need to live, help prevent the annoying sniffling and sneezing of allergies, boosting your immune system, helps to control high blood pressure and cholesterol, and certain types of injurious cancers. They can also be incorporated into food products (e.g. pastas, biscuits, bread, snack foods, candies, yoghurts, soft drinks), providing the health promoting affects that are associated with microalga biomass, probably related to a general immune-modulating effect.

Keywords: novel protein food sources, micro-organism, globalization, microbial biomass, genetic traits, plant breeding

Introduction
Food sources that can nourishes our body by providing most of the protein you need to live, help prevent the annoying sniffling and sneezing of allergies, boosting your immune system, helps to control high blood pressure and cholesterol, and certain types of injurious cancers. Does such a “designer food” exist in our surrounding that easily fulfils our body demands? Yes these are known as Novel protein food sources.
Novel Protein Foods (NPFs) are products based on vegetable protein and micro-organisms. A wide variety of vegetarian alternatives is already available on the market: seitan, tofu, soy meat, tempeh, quoin and meatless based on lupins. In addition, there are milk drinks that are not based on dairy (such as ‘soy milk’) and egg substitutes. The production of in vitro meat is proposed by some as a possible alternative. Insects, such as grasshoppers, crickets, caterpillars, beetles, ants, bees, wasps, termites, butterflies and moths, make up an integral part of the diet in large parts of the world. The consumption of seaweeds and algae has its origin in Asia, but the demand is growing [1].

Spirulina

Spirulina, filamentous blue-green microalgae or cyanobacteria, is well known as a source of protein consumed by humans and animals (60-70 g/100 g) of high biological value, since it is a rich source of vitamins, mainly vitamin B12 and pro-vitamin A, minerals, especially iron, and glinolenic acid, essential fatty acids precursor for prostaglandins [2]. Spirulina is the dried biomass of the cyan bacterium Arthrospira platensis, it has been widely used in several countries, it is considered GRAS (generally recognized as safe), without toxicological effects, and it is approved by the FDA (U.S.A.) and ANVISA[3] The protein efficiency ratio of Spirulina has been reported to be higher than vegetables, cereals and soy proteins. It is a complete protein containing all essential amino acids, though with reduced amounts of methionine, cysteine and lysine when compared to the proteins of meat, eggs and milk.

Kelly *et al*., (2011) [4] Spirulina is effective for victims of malnutrition diseases like kwashiorkor, where the ability of intestinal absorption has been damaged. Given to malnourished children, it is more effective than milk powders because milk's lactic acid can be difficult to absorb [4]. Simpore *et al*. (2005 and 2006) [5] suggested that spirulina may be a good food supplement for undernourished children. In particular, spirulina also seems to correct anemia and weight loss in HIV infected children, and even more quickly in HIV negative undernourished children [5]. Active ingredients of Spirulina are [6]. :

Table 1: Spirulina (dried) Nutritional value per 100 g

Nutrient Amount	Nutrient Amount
Energy 1,213KJ	Cystine 0.662g
Carbohydrates 23.9g	Phenylalanine 2.777g
Sugars 3.1g	Tyrosine 2.584g
Dietary fibre 3.6g	Valine 3.512g
Fat 7.72g	Arginine 4.147g
Saturated 2.65g	Histidine 1.085g
Monounsaturated 0.675g	Alanine 4.515g
Polyunsaturated 2.08g	Aspartic acid 5.793g
Protein 57.47g	Glutamic acid 8.386g
Tryptophan 0.929g	Glycine 3.099g
Threonine 2.97g	Proline 2.382g
Isoleucine 3.209g	Serine 2.998g
Leucine 4.947g	Phosphorus (17%) 118 mg
Lysine 3.025g	Potassium (29%) 1363 mg
Methionine 1.149g (4%) gμ	Sodium (70%) 1048 mg
Vitamin A equiv 29 beta-carotene (3%) gμ	Zinc (21%) 2 mg
lutein zeaxanthin 342 gμ0	Choline (13%) 66 mg
Thiamine(B1) (207%) 2.38 mg	Vitamin C (12%) 10.1 mg
Riboflavin (B2) (306%) 3.67 mg	Vitamin E (33%) 5 mg
Niacin (B3) (85%) 12.82 mg	Vitamin K (24%) gμ25.5
Pantothenic- (70%) acid (B5) 3.48 mg	Trace metals Calcium (12%)
Vitamin B6 (28%) 0.364 mg	120 mg
Folate (B9) (24%) gμ94	Iron (219%) 28.5 mg
Magnesium (55%) 195 mg	Other constituents
Manganese (90%) 1.9 mg	Water 4.68 g

g = micrograms, μ mg = milligrams, IU = International units Source: USDA Nutrient Database

Single-cell proteins (SCP)

Single-cell proteins refers to edible unicellular microorganisms. The biomass or protein extract from pure or mixed cultures of algae, yeasts, fungi or bacteria may be used as an ingredient or a substitute for protein-rich foods, and is suitable for human consumption or as animal feeds. Industrial agriculture is marked by a high water footprint, high land use, biodiversity destruction, general environmental degradation and contributes to climate change by emission of a third of all greenhouse gases, production of SCP does not necessarily exhibit any of these serious drawbacks. As of today, SCP is commonly grown on agricultural waste products, and as such inherits the ecological footprint and water footprint of industrial agriculture. However, SCP may also be produced entirely independent of agricultural waste products through autotrophic growth. Thanks to the high diversity of microbial metabolism, autotrophic SCP provides several different modes of growth, versatile options of nutrients recycling, and a substantially increased efficiency compared to crops.

Microorganisms

Microbes employed include

Yeast

- *Saccharomyces cerevisiae*
- *Pichia pastoris*
- *Candida utilis*
- *Torulopsis*
- *Geotrichum candidum*

Fungi (Mycoprotein)

- *Aspergillus oryzae*
- *Fusarium venenatum*
- *Sclerotium rolfsii*
- *Polyporus*
- *Trichoderma*
- *Scytalidium acidophilum*

Bacteria

- *Rhodobacter capsulatus*

Algae

- spirulina (dietary supplement)
- *Chlorella*

Advantages

Large-scale production of microbial biomass has many advantages over the traditional methods for producing proteins for food or feed.

Microorganisms have a much higher growth rate (algae: 2–6 hours, yeast: 1–3 hours, bacteria: 0.5–2 hours). This also allows selecting for strains with high yield and good nutritional composition quickly and easily compared to breeding. Whereas large parts of the crop, such as stems, leaves and roots are not edible, single-cell microorganisms can be used entirely. Whereas parts of the edible fraction of crops contains is indigestible, many microorganisms are digestible at a much higher fraction. Microorganisms usually have a much higher protein content of 30–70% in the dry mass than vegetables or grains. The amino acid profiles of many SCP microorganisms often have excellent nutritional quality, comparable to a hen's egg. Some microorganisms can build vitamins and nutrients which eukaryotic organisms such as plants cannot produce or not produce in significant amounts, including vitamin B12.Microorganisms can utilize a broad spectrum of raw materials as carbon sources including alkanes, methanol, methane, ethanol and sugars. What was considered "waste product" often can be reclaimed as nutrients and support growth of edible microorganisms? Like plants, autotrophic microorganisms are capable to grow on CO_2. Some of them, such as bacteria with the Wood–Ljungdahl pathway or the reductive TCA can fix CO2 between 2-3, up to 10 times more efficiently than plants when also considering the effects of photo inhibition. Some bacteria, such as several homo acetogenic clostridia are capable to perform syngas fermentation. This means they can metabolize synthesis gas, a gas mixture of CO, H_2 and CO_2 that can be made by gasification of residual intractable bio wastes such as lignocellulose. Some bacteria are diastrophic, i.e. they can fix N_2 from the air and are thus independent of chemical N-fertilizer, whose production, utilization and degradation causes' tremendous harm to the environment, deteriorates public health, and fosters climate change. Photosynthetic microorganisms can reach a higher solar-energy-conversion efficiency than plants, because in photo bioreactors supply of water, CO_2 and a balanced light distribution can be tightly controlled.

Yeast

Yeast single-cell protein (SCP) is a high- nutrient feed substitute. Among these, most popular are yeast species Candida, Hansenula, Pitchia, Torulopsis and Saccharomyces. The production of single cell protein using *Saccharomyces cerevisiae* grown on various fruits waste. The increasing world deficiency of protein is calling for greater demand of protein sources like India, although a developed nation, but its major population is facing nutrition deficiency and food scarcity problems. In the face of such worldwide issues, single cell proteins derived from the waste organic products had been proved a very useful technology. Dried cells of bacteria, algae, yeast, and fungi, which are rich in proteins and could be used as dietary supplements, are called Single Cell Proteins (SCP). Microbial protein or SCP has various benefits over animal and plant proteins in that its requirement for growth are neither seasonal or climate dependent; it can be produced all-round the year. It does not require a large expanse of land and it has high protein content with wide amino acid spectrum, low fat content and higher protein carbohydrate ratio than forages. It can be grown on waste and it is environmental friendly as it helps in recycling waste [7]. Yeast, a single cell prtein and have been recognized as noel protein sources as they are easy to harvest because of their size (larger than bacteria), containing high level of malic acid content, highlysine content,can grow at acidicpH, having long history of traditional use and high protein content (nearly about 60%) found in yeast. It is a significant source of some B-complex vitamins and contains trace amounts of several other vitamins and minerals also fortified with vitamin B.

Soy Protein Sources

Soy Protein isolates are also a much used protein source in lactose-free infant formulae for infants With lactose intolerance and/or galactosemia, and for vegan families. However, although the cow's milk protein is replaced by soy protein isolates, 10–14% of infants with cow's milk protein allergy are also allergic to soy protein, so it is better to use a hydrolysed protein formula. Furthermore, the effects of soy phytoestrogens on infants are still unclear [8].

Tempeh

Tempeh is a traditional Indonesian soy products that is made from fermented soybeans. A special fungus is used, which has the Latin name *Rhizopus oligosporus*, usually marketed under the name *Tempeh starter*. Tempeh's fermentation process and its retention of the whole bean give it a higher content of protein, dietary fibre, and vitamins. It has a firm texture and an earthy flavour, which becomes more pronounced as it ages.

Nutritional composition of tempeh

The soy carbohydrates in tempeh become more digestible as a result of the fermentation process.

In particular, the oligosaccharides associated with gas and indigestion is greatly reduced by the *Rhizopus* culture. Tempeh is also very high in fibre, offering more than a quarter of our daily fibre needs in one serving. As if the protein, fibre, and probiotic boosts weren't enough, tempeh is also a great source of many different vitamins and minerals. Tempeh is also an excellent source of magnesium, B vitamins, phosphorus, and manganese—key nutrients that promote a healthy sleep cycle.

It Contains Prebiotics

Tempeh is fermented using a fungus and is usually cooked before eating. Additionally, commercial products are pasteurized. However, tempeh seems to be rich in prebiotics -- types of fibre that promote the growth of beneficial bacteria in your digestive system.

It's High in Protein to Keep You Full

Tempeh is high in protein. One cup (166 grams) provides 31 grams of protein. Some studies suggest that a protein-rich diet may stimulate thermogenesis, leading to an increase in metabolism and helping your body burn more calories after each meal. A diet high in protein can also aid in appetite control by increasing fullness and decreasing hunger.

It May Reduce Cholesterol Levels
Tempeh is traditionally made from soybeans, which contain natural plant compounds called is flavones. Soy is flavones have been associated with reduced cholesterol levels. One review looked at 11 studies and found that soy is flavones were able to significantly decrease both total and LDL cholesterol.

Tofu
Tofu is very versatile as a food; it can be served fresh or cooked with vegetables and/or meat in thousands of different dishes and soups. It can also be further processed into various secondary tofu products, including deep-fried tofu, savoury tofu, grilled tofu, frozen tofu, dried-frozen tofu, fermented tofu, and etcetera. Tofu is made from soybean curds naturally gluten-free and low in calories. It contains no cholesterol and is an excellent source of iron and calcium. It also contains is flavones such as phytoestrogens [9]. These may help protect against cancers, heart disease, and osteoporosis. However, overconsumption may also present some risks. Here are some key points about tofu. Tofu is an important source of protein for many vegetarians and vegans. It may help lower "bad" LDL cholesterol. It may offer relief for certain symptoms of menopause.

Benefits
Tofu can be served instead of meat or incorporated into a variety of dishes. A diet that contains a variety of plant-based foods appears to contribute to overall health and wellbeing, and a lower risk of conditions such as obesity, diabetes, and heart disease. It can enhance the skin and hair, boost energy, and help maintain a healthy weight.

Cardiovascular disease
Soy oflavines have been found to help reduce levels of LDL "bad" cholesterol, although it does not seem to increase HDL or "good" cholesterol levels. Studies have indicated that daily consumption of soy may decrease markers for cardiovascular disease risk, including weight, body mass index (BMI), and total cholesterol. Consuming tofu as an alternative to animal protein can help lower levels of LDL cholesterol. This, in turn, decreases the risk of atherosclerosis and high blood pressure [10].

Breast and prostate cancer
Several clinical and experimental investigations have suggested that genistein, the predominant is flavone in soy, has antioxidant properties that may inhibit the growth of cancer cells.

Type 2 diabetes
People with type 2 diabetes often experience kidney disease, causing the body to excrete an excessive amount of protein in the urine. Evidence from one study has indicated that those who consumed only soy protein in their diet excreted less protein than those who only consumed animal protein. The researchers propose that this could benefit patients with type 2 diabetes.

Kidney function
Protein, and particularly soy protein, may enhance renal function, and it could have benefits for people who are undergoing dialysis or kidney transplantation. One meta-analysis of nine trials showed a positive effect of soy on some biomarkers of those with chronic kidney disease [11]. This may be due to its protein content, but also because of its impact on lipid levels in the blood.

Osteoporosis
Soy is flavones may help reduce bone loss and increase bone mineral density, especially after menopause. They have also been reported to reduce some other symptoms of menopause.

Liver damage
One study in rats has suggested that any type of tofu that has been curdled with various coagulants may help prevent liver damage caused by free radicals.

Age-related brain diseases
Population studies have indicated that, in regions where people consume more soy, there is a lower incidence of age-related mental disorders. When the same group carried out a further small study, involving 65 people over the age of 60 years with Alzheimer's, they did not find that soy is oflavines offered any cognitive benefits. However, findings published in 2017 suggested that soy products may help people with Alzheimer's due to their lecithin content, which helps the body produce the phospholipids phosphatides acid (PA) and phosphatidylserine (PS). PA and PS play an important role in the functioning of neurones [12].

Textured vegetable proteins
Textured vegetable proteins have a PER (protein efficiency ratio) of at least 80% of casein. Soy protein contains all the essential amino acids. The amino acid lysine is present in substantial levels, and methionine is the first limiting amino acid. Methionine may be added to soy protein products to give a PER value equal to casein. The processes involved in fabricating textured vegetable proteins provide an opportunity to incorporate essential nutrients into food products. Advantages of textured vegetable proteins use in canned food products include: The extruded products are dried to less than 8% moisture and under normal storage conditions have a shelf-life of about one year. Freezers or cold storage are not required. Textured vegetable proteins can be fortified with vitamins, minerals, and other supplements to provide balanced nutrition in the final canned product. Textured vegetable proteins maintain their structure upon hydration and provide a meat-like texture. They normally absorb two to three times their weight in water and have good fat-absorption properties. The products can be coloured, flavoured, and sized to resemble a wide variety of food products. They allow canners a means of making meat-like canned products in the off-season, thereby allowing year-around utilisation of equipment.

Mycoprotein
Mycoprotein is a form of single-cell protein, also known as fungal protein. Fungi have been influencing human affairs for thousands of years, whether as a direct food source, as a medicine, or in a food. *Fusarium venenatum*, the principal ingredient of Mycoprotein is an Ascomycota, one of the largest groups within the fungi family, which also includes truffles and morels. It is one of a genus of filamentous fungi, meaning it is comprised of a web of finely spun strands (hyphae). Fungi have provided food for man, primarily in the

form of fruit bodies of basidiomycetes and/or ascomycetes [13].

Nutritional value

Mycoprotein contains all nine essential amino acids – meaning the body cannot produce them, and so must be obtained through the diet – making it a high-quality protein, comparable to protein sources such as meat or fish. The nutritional value of Mycoprotein, is found to be comparable with eggs in amino acid composition. Mushrooms are one of the most common fungi which can be used as the source of protein as they have the high protein content, usually around 20-30% by dry weight. Mycoprotein is high in protein, high in fibre, low in saturated fat, and contains no cholesterol. This meat-free protein contributes to a balanced plant-based eating style.

Leaf Protein Extract

Leaf protein concentrate (LPC) is a concentrated form of the proteins found in the leaves of plants. It has been examined as a human or animal food source, because it is potentially the cheapest, most abundant source of available protein. Although humans can derive some protein from the direct consumption of leaves as leaf vegetables, the human digestive system would not be able to deal with the enormous bulk of leaves needed to meet dietary protein requirements with leaf vegetables alone [14].

Nutritional value

The protein content of the dried product from the sources is about 50-70%. Leaf protein concentrate (LPC) are very nutritious food made by separating mechanically indigestible fibre and soluble anti-nutrients from much of the protein, vitamins, and minerals in certain fresh green plant leaves. They are rich in β-carotene, iron, and high-quality protein so it will be very effective in fighting malnutrition. The amino acid profile of leaf protein concentrate indicates that it is nutritionally superior to most cereal and legume seed proteins; it can also compare positively with animal proteins except egg and milk.

Duckweed: An Innovative Protein Source

The family of duckweeds (Lemnaceae family) flourish, floating atop still or slow-moving fresh water in most parts of the globe. Their reproduction can be extremely rapid, doubling mass in as few as two days, forming a virtual carpet across the water. Out of about 40 species of duckweed, the most prominent is Lemma minor [15]. Protein content of the dried plant is in the range of 25 percent to 35 percent 1.The amino acid profile of its protein concentrate meets the World Health Organization's (WHO) recommendations for amino acids, except for methionine. Duckweed is rich in lysine, which makes it worthy as a "complementary" protein in low-lysine vegetarian diets based on rice or corn. Analysis has indicated duckweed species also contain a good number of branched-chain amino acids (BCAAs [valine, leucine and isoleucine]), which are sought-after in products designed for athletes. And, duckweeds have good content of omega-3 fatty acids, resulting in a favourable omega-6: omega-3 ratio of 0.5 or less. Duckweed species therefore require careful cultivation, grooming and management for use in human food and supplement products. For optimum production, duckweed needs to be monitored, grown on surface partitions that are shielded from wind, maintained at a favourable mass by fertilization to adjust nutrients, and harvested to retain peak yields [16].

Seaweed

Researchers have previously shown that protein-rich red seaweeds such as Palmariapalmata (common name Dulse) and Porphyra (common name Sleabhac or Laver) species may potentially be used in the development of low-cost, highly nutritive diets that may compete with current protein crop sources such as soya bean. Researchers have found that some of these seaweed proteins may have health benefits beyond those of basic human nutrition -- for use in functional foods. Bioactive peptides are food-derived peptides that exert a physiological, 'hormone-like', beneficial health effect. Proteins and peptides from food sources such as dairy, eggs, meat and fish are well documented as agents capable of reducing high blood pressure and are thought to be able to prevent CVD.

MICRO ALGAE-

Microalgae are an enormous biological resource, representing one of the most promising sources for new products and applications. They can be used to enhance the nutritional value of food and animal feed, due to their well-balanced chemical composition. Moreover, they are cultivated as a source of highly valuable molecules such as polyunsaturated fatty acids, pigments, antioxidants, pharmaceuticals and other biologically active compounds. The application of microalgal biomass and/or metabolites is an interesting and innovative approach for the development of healthier food products. Microalgae use by indigenous populations has occurred for centuries. However, the cultivation of microalgae is only a few decades old and among the 30000 species that are believed to exist, only a few thousands strains are kept in collections, a few hundred are investigated for chemical content and just a handful are cultivated in industrial quantities.

Chlorella Vulgaris

Microalgae in Novel Food Products Chlorella vulgaris has been used as an alternative medicine in the Far East since ancient times and it is known as a traditional food in the Orient. It is widely produced and marketed as a food supplement in many countries, including China, Japan, Europe and the US, despite not possessing GRAS status. Chlorella is being considered as a potential source of a wide spectrum of nutrients (e.g. carotenoids, vitamins, minerals) being widely used in the healthy food market as well as for animal feed and aquaculture. Chlorella is important as a health promoting factor on many kinds of disorders such as gastric ulcers, wounds, constipation, anaemia, hypertension, diabetes infant malnutrition and neurosis.

It is also attributed a preventive action against atherosclerosis and hypercholesterolemia by glycolipids and phospholipids, and antitumor actions by glicoproteins, peptides and nucleotides.

Haematococcus Pluvial is

Haematococcus pluvial is has been identified as the organism which can accumulate the highest level of astaxanthin in nature (1.5-3.0% dry weight). This carotenoid pigment is a potent radical scavenger and singlet oxygen quencher, with increasing amount of evidence suggesting that surpasses the antioxidant benefits of b-carotene, vitamin C and vitamin E. Haematococcus is currently the prime natural source of this

pigment for commercial exploitation, particularly in aquaculture salmon and trout farming (Lorenz and Cysewski, 2000) [19].

Table 1. Major microalgae commercialized for human nutrition (Adapted from Pulz and Gross, 2004, Spolaore *et al*., 2006 and Hallmann, 2007) [18]

Microalga	Major Producers	Products	World production (t/year)
Spirulina (Arthrosphira)	Hainan SimaiPharmacy Co. (China)	powders, extracts tablets, powders, xtractstablets, powders, beverages, extractstablets, chips, pasta and liquid extract	3000
Chlorella	Taiwan Chlorella Manufacturing Co. (Taiwan) Klötze (Germany)	tablets, powders, nectar, noodlespowders	2000
Dunaliella salina	Cognis Nutrition and Health (Australia)	Powders b-carotene	1200
Aphanizomenon flos-aquae	Blue Green Foods (USA) Vision (USA)	capsules, crystals powder, capsules, crystals	500

Conclusion

The aspire of this paper is to explore and review the state of the nutritional profile of major novel protein sources for the future perspective as they become the alternative sources of animal protein particular spirullina, insects, rapeseed, duckweed, microalgae, and seaweed as the demand of novel food sources become increasing day by day because of International travel, Globalization, Changing Tastes, Increasing Desire to taste new food instead of traditional one, Advances in genetics traits, Innovation in field of plant breeding, Rapid increasing population, Increasing demands of food resources, Problem of under-nutrition.

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