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Antioxidant properties and utilization of Spirulina by different processing methods

Neeraj Verma and Sunita Mishra

Abstract
The utilization of spirulina powder is a good source of potentially valuable for food, pharmaceutical nutraceutical and supplements industries. The using of spirulina as an antioxidant in food product is neutralizing the free radicals generated due to oxidative stress. Food preservation and processing become vital in order to utilized and maintain its organoleptic properties like taste, flavor, and appearance. It is a great source to utilize the spirulina by the processing of different product formation as spirulina soup sticks, spirulina soup and spirulina pizza base. The aim of this present review is to summarize the current information about the utilization of spirulina to use as an antioxidant and supplements and enhance the nutritive properties which can be used as food fortification.

Keywords: spirulina, antioxidant, supplements, nutraceutical, fortification

Introduction
Spirulina is one of the most widely identified species of algae and is an excellent source of essential amino acids, fat containing PUFA, minerals and vitamins (Rajesha *et al.*, 2009) [12]. US department of agriculture considered "Spirulina as the most ideal food for mankind" and regarded it as "Food for the future" in 1988. The abundance of bioactive substances in Spirulina is of great importance from nutritional point of view. Thus, the cyanobacterial biomass provides new opportunity for manufacture of functional dairy foods (Varga *et al.*, 2002) [13] and also bakery foods. Spirulina represents considerable potential to be used in food industry attributed to their versatile nutritional.

The dried cells of microorganisms such as bacteria, fungi, yeasts and algae that are grown in large-scale culture systems as proteins, for human or animal consumption are collectively known as single cell proteins (Sasson, 1997) [2]. Single cell proteins (SCP) are characterized by fast growth rate, high protein content (43-85%) compared to field crops, require less water, land and independent of climate, grow on wastewater, can be genetically modified for desirable characters such as amino acid composition and temperature tolerance (Tri-Panji and Suharyanto. 2001) [1]. Microorganisms used to produce biomass and the choice of a microorganism depends on numerous criteria, the most important of which is the nature of the raw material available. The ideal microorganism should possess the following technological characteristics like high specific growth rate, biomass yield, low nutritional requirements, ability to develop high cell density, stability during multiplication, capacity for genetic modification and good tolerance to temperature and pH. In addition, it should have a balanced protein and lipid composition. It must have a low nucleic acid content, good digestibility and to be non-toxic.

Nowadays, food industries need new food ingredients obtained from natural sources and developed novel functional foods or nutraceuticals. Microalgae now combine the traditional and new biotechnologies where microalgal biomass can be used as a source of proteins, biochemicals, lipids, polysaccharides and colorants (Athukorala, 2006) [3]. Spirulina (Arthrospira) is blue-green algae found in alkaline Lakes around the world. The name "Spirulina" is derived from the Latin word for "helix" or "spiral", referring to the physical structure of the organism. It is motile multicellular filamentous blue-green algae and reproduces by binary fission. Spirulina is a non-nitrogen-fixing blue-green alga and cell wall made of mucopolysaccharide its soft and easily digestible nature, which makes it safe for human consumption. Spirulina is capable of growing in high alkalinity with the presence of carbonate, bicarbonates and inorganic nitrogen (Aiba and Ogawa, 1977) [4]. The ability of Spirulina to grow in hot and alkaline environments ensures its hygienic status, as no other organisms can survive to pollute the waters in which this alga thrives. Spirulina is one of the cleanest, most naturally sterile foods found in nature. It has been used as feed for fish, poultry and farm animals (Tragut *et al.*, 1995; Abdulqader *et al.*, 2000) [5, 6].

Microalgal species are an important alternative material to extract natural antioxidant compounds to delay or prevent the oxidative damage caused by Reactive Oxygen Species (ROS) molecules (El-Baz *et al.*, 2002) [7]. Spirulina has reported to prevent oxidative damage and hence can indirectly reduce cancer formation in human body. In this respect, the increased consumption of foods characterized by free radical scavenging activity, leads up to a doubling of protection against many common types of cancer formation (Cooke *et al.*, 2002; Romay *et al.*, 2003; Anbarasan *et al.*, 2011) [9, 8].

Table 1: General Composition of Spirulina [12]

Protein	60 % - 69 %
Carbohydrates	16 % - 20 %
Lipids	5% - 7 %
Minerals	6 % - 9%
Moisture	2.5% - 6.0%

Nutrient profile of spirulina vs other foods

- 180% more calcium than whole milk.
- 670% more protein than tofu.
- 3100% more beta carotene than carrots.
- 5100% more iron than spinach.
- more antioxidant and anti-inflammatory.
- activity in 3g of Spirulina than in five servings of fruits and vegetables. (Moorhead K *et al*., 2005) [10]

Spirulina as antioxidant

One of most important characteristics of Spirulina is its antioxidant property. Antioxidants are the substances which neutralize the free radicals generated due to oxidative stress. Free radicals are not only nuisance molecules in the atmosphere but can cause havoc inside the body. They are the unstable products of normal cell processes. Free radicals can damage the concerned cell & lead to the death of these cells. Oxidative stress directly or indirectly leads to various disorders like diabetes, atherosclerosis, rheumatoid arthritis, recurrent aphthous stomatitis, cancer, etc. Very high amounts of beta-carotene, tocopherol and combined form of these antioxidants make Spirulina a very good source of natural antioxidant along with high protein.

These antioxidants can become pro-oxidants and protect the body from oxidative stress. (Arpita Mohan *et al*., 2014) [11].

Materials and Methods

Research design

Research design is a coherent plan in conducting research which deals with investigation so conceived to obtain sample to research. Research design is used to conduct research objectivity of accuracy. The research design followed in the present study by research on spirulina and their products available in the market. Spirulina is use as supplements, not using in daily food. The research was developing innovative food products by using of spirulina in daily food, especially for sports persons.

Locate of study

The study was conducted in the laboratory of the Department of Food Science and Technology, School for Home science, BBAU, Lucknow and the analysis laboratory of RFRAC (Regional Food Research and Analysis Centre) located in Lucknow.

Period of the study: The present study was carried out 6 months during session 2017-2018 between January 2018 to May 2018.

Study design: The approach for this study was purposively one. To developed the products.

Sample size

The sample size is based on three different products prepared by spirulina. These products are soup, soup sticks and pizza base. All these products were different ratio of spirulina. Products are prepared with spirulina and other ingredients.

Table 2: Ingredients preparation for spirulina soup

Ingredients	Amount
Refine flour	200g
Corn flour	200g
Soy flour	200g
Wheat grass powder	200g
Moong dal flour	100g
Flaxseed (roasted)	50g
Spirulina powder	20g
Salt	2 tsp.

Table 3: Ingredients preparation for soup sticks

Ingredients	Amounts
Refine flour	250g
Flaxseed (soaked)	20g
Spirulina	20g
Salt	1tsp.
Fresh active yeast	1 tsp.
Sugar	½ tsp.

Table 4: Ingredients preparation for pizza base

Ingredients	Amounts
Refine flour	200g
Spirulina	20g
Fresh active yeast	1tsp.

Sampling design

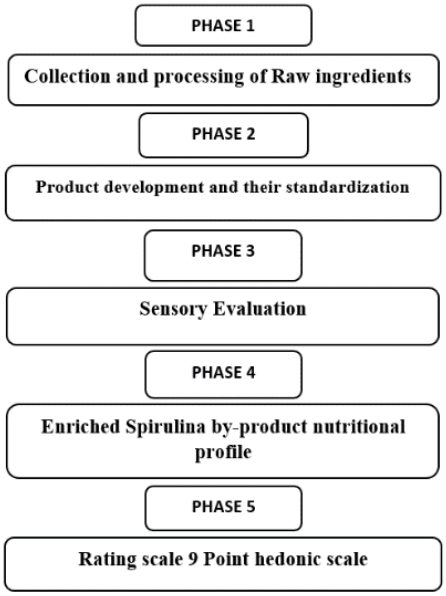

Fig 1: Flow chart for manufacturing of Spirulina by-products

Preparation of spirulina soup powder

Step 1. Measuring all ingredients
Step 2. Add all ingredients
Step 3. Mixing well all dry form
Step 4. Collect whole ingredients mixer and giving heat treatment at 60˚c for 5 – 10 minutes.
Step 5. Final product spirulina soup powder.
Step 6. Prepared spirulina soup ready to eat (Mix with hot water).

Preparation of spirulina soup sticks

Step 1. Use fresh yeast (Activated).
Step 2. Measuring refines flour and flaxseed.
Step 3. Mix all ingredients (Refine flour, flaxseeds, yeast, black salt, spirulina and add water or some butter/ oil for Greasing).
Step 4. Prepared dough and keep it for proofing for 30 minutes.
Step 5. After proofing give the shape and cut into small pieces.
Step 6. Roll with hand, place it on oven trey.
Step 7. Preheated oven at 150˚c, place the trey in the oven at 180˚c 30-45 minutes. prepared spirulina soup sticks ready to eat.

Preparation of spirulina pizza base.

Ingredients- Refine flour, spirulina and active fresh yeast.
Step 1. Active fresh yeast
Step 2. Collect all ingredients, prepared dough.
Step 3. Weighing dough and cut into desirable pieces
Step 4. Keep dough for proofing, for 30 minutes.
Step 5. After proofing, punch it and keep it again for proofing for 30 minutes
Step 6. Prepare pizza base on baking tray, Pizza base half bake at 180˚c for 20 – 30 minutes.
Step 7. For preparing pizza take vegetable, pizza Toppin and mozzarella cheese. Speared on base than bake again at 150˚c for 20 minutes.

Results and Discussion

Sensory evaluation of packed and processed spirulina by products by experts' panel of members on hedonic scale and marking was done on the six parameters-

- Colour
- Body and texture
- Flavour
- Appearance
- Taste
- Overall acceptability

Treatment

The experimental spirulina by products (Spirulina soup, spirulina soup sticks and spirulina pizza base) of spices were characterized as developed product in the present study. The various parameters were incorporated for product development to reach acceptability and human population. For that sensory evaluation process was done by of panellist constitute 5 Members in the expertise field of nutrition.

Parameter 1: Colour Quality

Table 5: Individual marking for colour

Member	T1	T2	T3
1	7	8	7
2	7	7	8
3	6	8	7
4	8	8	8
5	7	6	8
Total	35	37	38

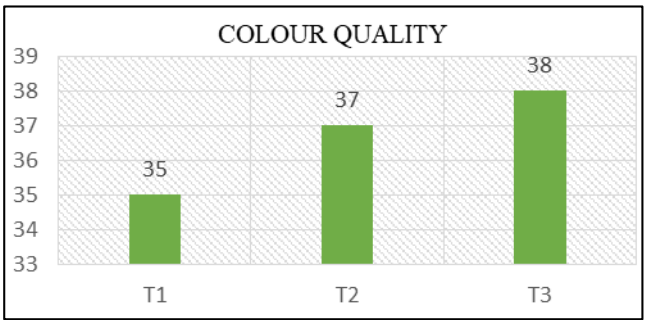

Fig 2: Graphical representation of colour

Above graph shows that T3 is most acceptable sample T3 in term of colour among the sensory panellist members and it get highest scoring. Then after sample T2 and sample T1 respectively.

Parameter 2: Body and texture

Table 6: Individual marking for body and texture

Member	T1	T2	T3
1	6	7	7
2	8	8	8
3	7	6	8
4	8	8	7
5	7	6	8
Total	36	35	39

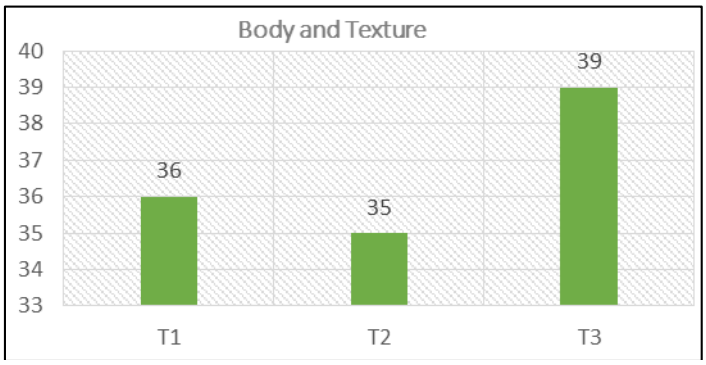

Fig 3: Graphical Representation - Body and Texture

Above graph shows that T3 is most acceptable sample T3 in term of texture among the sensory panellist members and it get highest scoring. Then after sample T1 and sample T2.

Parameter 3- Flavour

Table 7: Individual marking for flavour

Member	T1	T2	T3
1	8	7	7
2	8	6	8
3	8	6	7

4	7	8	7
5	8	6	8
Total	39	33	37

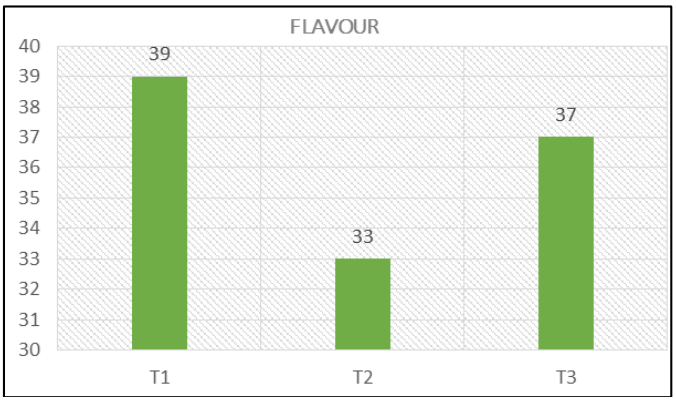

Fig 4: Graphical representation flavour

From the graph, it shows that T1 is most acceptable in term of flavour among the sensory panellist members and it get highest scoring. Then after sample T3 and sample T2.

Parameter 4 - Appearance

Table 8: Individual marking for appearance

Member	T1	T2	T3
1	8	7	7
2	8	8	8
3	7	7	6
4	7	7	6
5	8	6	8
Total	38	35	35

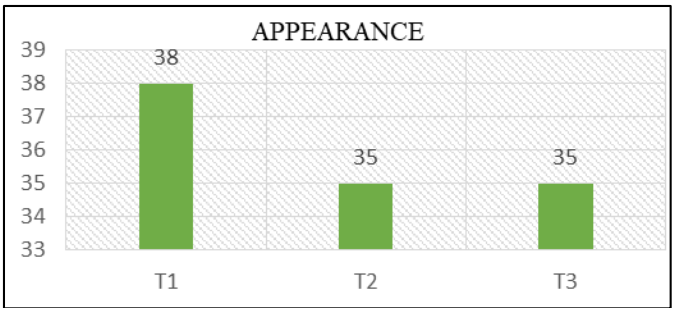

Fig 5: Graphical representation – appearance

From the graph, it shows that T1 is most acceptable sample T1 in term of appearance among the sensory panellist members and it get highest scoring. Then after sample T2 and sample T3 are equal.

Parameter 5 - Taste

Table 9: Individual marking for taste

Member	T1	T2	T3
1	7	7	7
2	6	8	8
3	7	6	7
4	6	7	7
5	8	7	7
Total	34	35	35

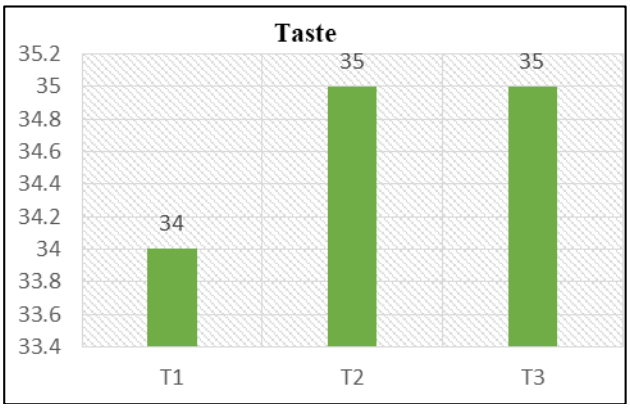

Fig 6: Graphical Representation – Taste

From this graph, it shows that sample T2 and T3 is equally acceptable in term of taste among the sensory panellist members and both samples are get same scoring. Then after sample T1 respectively.

Table 10 - Overall acceptability

Overall calculation is done to know most acceptability of the product in terms of quality by sensory evaluation scoring given by the panellist Members, in this all scoring of colour, flavour consistency and absence of defects are calculated in the table, by this get do statistical analysis and obtained standard deviation, average and other calculation.

Table 10: Overall acceptability

Member	P1	P2	P3	P4	P5	Total	Average	SD
1	34	38	39	36	35	182	36.4	2.074
2	35	35	33	35	37	175	35	1.414
3	35	35	37	39	38	184	36.8	1.788

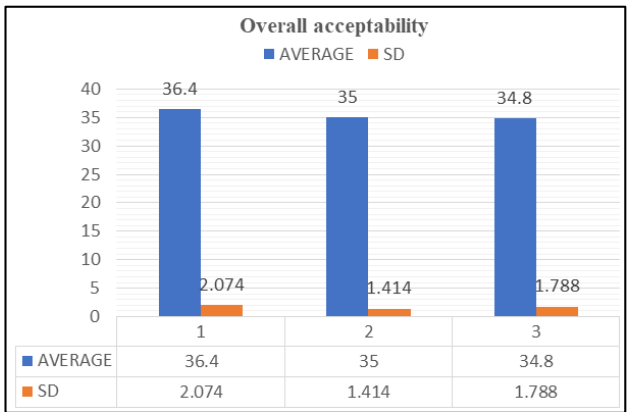

Fig 7: Graphical representation of average score and standard deviation for overall calculation

Overall calculation

- In this table of overall calculation, we got the average of T1, T2 and T3 are 36.4, 35 and 34.8 respectively.
- Standard deviation is 2.074, 1.414 and 1.788 respectively.

Conclusion

The utilization of spirulina by- products enhances due to the quality of spirulina by products that are rich source of many utilizable component. The product formation by using the

spirulina by-products contain many health enhancement substances for supplement and balance diet. These products optimize the availability of antioxidant, protein, carbohydrate, minerals and fat. These foods are ready to eat which is very easily too consumed by others. For the insurance product quality, organoleptic indicator is good to determine the quality and freshness of product. The organoleptic evaluation of the spirulina by-product was done by using nine-point hedonic scales by panels of 5 members. The scoring for each of the product was done according to various parameters i.e. texture, taste, flavor, colour and appearance and overall acceptability.

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