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BIOLOGICAL EVALUATION OF ARTHROSPIRA PLATENSIS(SPIRULINA)

Botany

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ABSTRACT

Limited consumption of natural foods in the 21st century has led to deficiencies in essential vitamins and minerals in the human body. Excessive and repetitive use of chemical fertilizers has resulted in declining crop productivity. *Spirulina*, a multi-cellular and filamentous blue-green algae, is proposed as a viable solution to these problems. *Spirulina* is highlighted for its high content of macro and micronutrients, essential amino acids, proteins, lipids, vitamins, minerals, and antioxidants. It is described as one of the most nutritious and concentrated whole food sources, earning it the label of a "superfood." *Spirulina* exhibits various health-promoting properties, including anticancer, antidiabetic, anti-inflammatory, and immunomodulatory effects. *Spirulina* has the potential to mitigate the negative impacts of wastewater discharge through bioremediation. In countries like India where malnutrition is a significant social challenge, *Spirulina* supplementation in the diet is proposed as a means to combat malnutrition effectively. *Spirulina* has garnered significant attention from both research and industrial perspectives as a valuable source of nutraceuticals and pharmaceuticals. The present study aims to evaluate the secondary metabolites of *Spirulina*, specifically focusing on phenolic content and antioxidant activity using various assays such as Phosphomolybdenum activity, Metal chelating activity, and Superoxide radical scavenging activity. The study also reports the protein content of *Arthrospira platensis*as 25.76 mg/g. It also revealed the presence antioxidant properties by Phosphomolybdenum (164.88 mg AAE/g extract), Metal chelating activity (164.88 mg EDTA/g extract) Superoxide radical scavenging activity (37.86%,). *Spirulina* is seen as a cost-effective means of improving livestock and crop productivity in a sustainable manner, thus contributing to food and nutritional security. *Spirulina* has multifaceted potential to address key challenges in nutrition, agriculture, and environmental sustainability, making it a promising solution for a range of global issues.

KEYWORDS

Content, Antioxidant Properties, Environmental Sustainability, Superfood

INTRODUCTION

Spirulina is a single-cell protein rich in all essential nutrients and vitamins and can be used to produce functional food. In fact, one of the most important problems in the food industry is the use of synthetic food additives that increase the risk of cancer. Therefore, efforts are being made around the world today to isolate new and safe antioxidants from natural sources. Among these, the natural products of cyanobacteria are an important source of new drug compounds. Natural bioactive products not only have medicinal value themselves but are also used as building models to create synthetic analogs. Undernutrition is the outcome of insufficient food intake, resulting in a decadent nutritional status characterized by lower weight and/or height that those expected for one's age, UNICEF, 2006. Such condition, being frequently related to protein deficiency, constitutes a public health problem all over the world, but particularly in developing countries, Simpore et al., 2005.In the interest of developing novel more effective protein sources for preventing/reversing malnutrition, increasing attention has been turned to microalgae. The possible utilization of algae as a nonconventional protein source was suggested some decades ago (Belay et al., 1996)

Spirulina is of great interest as it offers the possibility of being used as a functional food, Ambrosi et al., 2008. This term refers to those foods that have proven to aid specific body functions, yielding health-promoting properties and reduce the risk of disease beyond its nutritional functions Hasler, 1996. Moreover, *Spirulina* has also proven to have good acceptance as of its organoleptic properties (thus making it a possible prospect for food or a nutrition supplement) and it has not exhibited neither acute nor chronic toxicities, making it safe for human consumption Chamaro et al.,1996;Salazar et al.,1998. The objective of the present paper was to analyse the total phenols and the antioxidant activity. This analysis aims to understand the phenolic content present in *Spirulina* and to evaluate its potential antioxidant properties. Total phenols are a group of compounds found in plants that have been extensively studied for their health benefits, particularly their antioxidant properties. By quantifying the total phenolic content and assessing the antioxidant activity, to help gain insights into the potential health-promoting effects of the sample being studied. This analysis is often conducted using various biochemical assays and analytical techniques to measure both the concentration of phenolic compounds and their ability to scavenge free radicals or inhibit oxidative processes. The results of this analysis can provide valuable information for various fields including food science, medicine, and natural product research

MATERIALS AND METHODS

Determination of the total protein in tissue homogenate

Protein content in the sample was determined according to the method of Lowry et al. (1951).The tyrosine and tryptophan residues of proteins cause reduction of the phosphomolybdate and phosphotungstate components of Folin-Ciocalteau reagent in an alkaline medium to give a bluish-purple colour with absorbance at 660 nm.About 10 µL of the homogenate was mixed with 990 µL of distilled water, 5 mL of alkaline CuSO4 (0.5% CuSO4 in 1% sodium potassium tartrate and 2% Na2CO3 in 0.1 N NaOH mixed in the ratio 1:50) was kept for 10 min at room temperature. Then 0.5 mL of 1 N Folin-Ciocalteau reagent was added and absorbance was measured after 30 min at 660 nm against the reagent blank. Protein content was calculated from the standard graph plotted using different concentrations (20-200 µg/mL) of bovine serum albumin (BSA).

Phosphomolybdenum assay

The antioxidant activity of *Spirulina* was evaluated by the green phosphomolybdenum complex formation according to the method of Prieto et al. (1999). An aliquot of 40 µL of sample or ascorbic acid in 1 mM dimethyl sulphoxide (standard) or distilled water (blank) was added with 1 mL of reagent solution (0.6 M sulphuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate) in a test tube. The test tubes were covered with foil and incubated in a water bath at 95°C for 90 min. After the samples had cooled to room temperature, the absorbance of the mixture was measured at 695 nm against the reagent blank. The results reported (Total antioxidant capacity) are mean values expressed as mg AAE/g extract (Ascorbic Acid Equivalents per gram extract).

Metal chelating activity

The chelating of ferrous ions by various extracts of *Spirulina* was estimated by the method of Dinis et al. (1994). Initially, about 100 µL the extract samples were added to 50 µL of 2 mM FeCl2 solution. Then the reaction was initiated by the addition of 200 µL of 5 mM ferrozine and the test tubes were vortexed well and left standing at room temperature for 10 minutes. The reaction mixture containing deionized water in place of sample was considered as the negative control. Absorbance of the solution was then measured spectrophotometrically at 562 nm against the blank (deionized water). EDTA was used as the standard metal chelating agent and the results were expressed as mg EDTA equivalents/ g extract.

Superoxide radical scavenging activity

The assay was based on the capacity of various extracts to inhibit formazan formation by scavenging the superoxide radicals generated in riboflavin–light–NBT system (Beauchamp and Fridovich, 1971).

Each 3 mL reaction mixture contained 50 mM sodium phosphate buffer (pH-7.6), 20 µg riboflavin, 12 mM EDTA, 0.1 mg NBT and 40 µL of aliquot of sample solution or BHA and BHT (standard). Reaction was started by illuminating the reaction mixture with sample extract for 90 s. Immediately after illumination; the absorbance was measured at 590 nm against the reagent blank (reaction mixture without plant sample). Identical tubes with reaction mixture kept in the dark served as negative control. The scavenging activity on superoxide anion generation was calculated as:

Scavenging activity (%) = [(A0 - A1) / A0] X 100

Where,

A0 is the absorbance of the control, and

A1 is the absorbance of the sample extract/standard.

RESULT

Protein analysis of *Arthrospira platensis* revealed 25.76 mg/ g (Table 1). A. platensis contains 60-71% of protein (Jongkon et al.,2008) or 55-70% based on Jung et al.,2019 compared to other sources such as dry soybeans (35%), grains (8-10%) or peanuts (25%) protein. These values make *Spirulina* essential for functional diet. Protein is needed for cell metabolism processes to support growth, which affects the synthesis process and accumulation of cell contents such as carbohydrates, amino acids, nucleic acids, and fats (Tokusoglu & Unal , 2006) .The protein in *Spirulina* is highly digestible and bioavailable, meaning that the body can efficiently absorb and utilize the protein it provides. In addition to protein, *Spirulina* is packed with other essential nutrients such as vitamins, minerals, antioxidants, and essential fatty acids. This makes it a highly nutritious food source, offering a wide range of health benefits beyond just protein content.Numerous scientific studies have examined the protein content and nutritional profile of *Spirulina*, consistently confirming its status as a rich protein source. These studies have also investigated its potential health benefits, including its role in supporting muscle growth, weight management, and overall health

Table 1. Total protein of *Arthrospira platensis*

Component	Sample
Total protein (mg/ g)	25.76 ± 0.41

Values are mean of triplicate determination (n=3) ± standard deviation

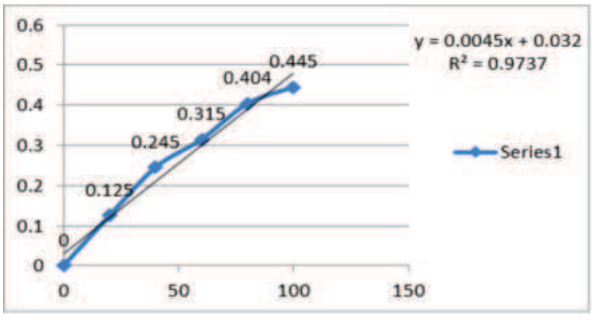

Fig. 1. Determination of the total protein of *Arthrospira platensis* ethanolic extract

Table 2 revealed the presence antioxidant properties by Phosphomolybdenum (164.88 mg AAE/g extract). In the Phosphomolybdenum assay, the antioxidant activity of a sample is measured based on its ability to reduce Mo(VI) to Mo(V). This reduction reaction produces a green phosphate/Mo(V) complex, the intensity of which is directly proportional to the total antioxidant capacity of the sample.

The value of 164.88 mg AAE/g extract indicates the antioxidant capacity of the sample in terms of ascorbic acid equivalents (AAE) per gram of extract. This suggests that the sample possesses significant antioxidant activity, which can help neutralize harmful free radicals and protect against oxidative stress-related damage.Overall, the results from Table 2 provide quantitative evidence of the antioxidant properties of the analyzed sample, highlighting its potential health-promoting benefits.

Table 2: Phosphomolybdenum assay of *Arthrospira platensis* ethanolic extract

Component	Sample
Phosphomolybdenum mg AAE/g (extract)	164.88 ± 0.96

AAE – Ascorbic Acid Equivalents

Values are mean of triplicate determination (n=3) ± standard deviation.

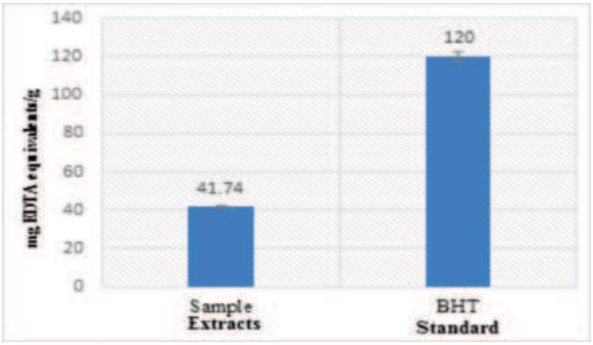

Fig. 3. Metal chelating activity of *Arthrospira platensis* ethanolic extract

Table 3 indicates the presence of antioxidant properties as determined by Metal Chelating Activity, with a value of 164.88 mg EDTA/g extract. Metal chelating activity refers to the ability of a substance to bind to metal ions, thereby preventing them from catalyzing oxidation reactions in the body.

This activity is an important aspect of antioxidant behaviour, as metals such as iron and copper can participate in free radical generation and oxidative stress when present in excess. The value of 164.88 mg EDTA/g extract suggests the chelating capacity of the sample in terms of ethylene diamine tetra acetic acid (EDTA) equivalents per gram of extract.

EDTA is a well-known chelating agent often used in laboratory assays to evaluate metal chelating properties. A higher metal chelating activity value indicates a greater ability of the sample to chelate or bind metal ions, thereby reducing their availability to catalyze oxidative reactions. This property is indicative of the potential of the sample to mitigate oxidative stress and protect against oxidative damage associated with various diseases.

In summary, the results from Table 2 provide quantitative evidence of the antioxidant properties of the analyzed sample, particularly in terms of its metal chelating activity, which may contribute to its overall health-promoting effects.

Table 3. Metal chelating activity of *Arthrospira platensis* ethanolic extract

Component	Sample	BHT
Metal chelating activity mg EDTA equivalents/g	164.88 ± 0.96	120 ± 2.4

Table 4 indicates the presence of antioxidant properties as determined by Superoxide Radical Scavenging Activity, with a value of 37.86%. Superoxide radicals are highly reactive oxygen species that can cause oxidative damage to cells and tissues if not neutralized. Therefore, the ability of a substance to scavenge or neutralize superoxide radicals is an important aspect of its antioxidant activity.

The value of 37.86% represents the percentage of superoxide radicals scavenged by the sample. In other words, this indicates the efficiency of the sample in neutralizing superoxide radicals, with a higher percentage indicating greater antioxidant activity.

Superoxide radical scavenging activity is typically measured using specific assays that assess the ability of a substance to inhibit the generation or react directly with superoxide radicals.

Overall, the results from Table 4 provide quantitative evidence of the antioxidant properties of the analyzed sample, particularly in terms of its superoxide radical scavenging activity. This suggests that the sample has the potential to mitigate oxidative stress and protect against cellular damage cased by superoxide radicals.

Table 4. Superoxide radical scavenging activity of *Arthrospira platensis* ethanolic extract

Component	Sample	BHA	BHT
Superoxide % of inhibition	37.86 ± 1.74	54.7 ± 0.25	54.2 ± 0.1

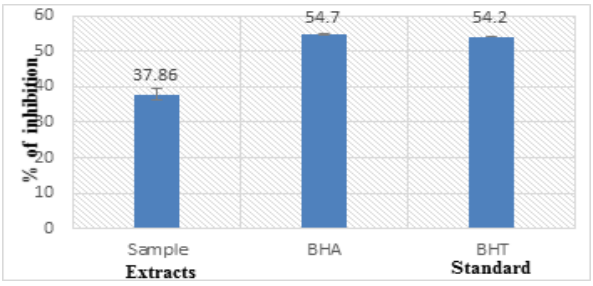

Fig 4. Superoxide radical scavenging activity of *Arthrospira platensis* ethanolic extract

CONCLUSION

The outcomes of this study suggest the potential use of *Arthrospira platensis* biomass as a sustainable source of protein for developing highly nutritional foods, especially when combined with other plant-based materials. *Spirulina* can indeed be categorized as a superfood due to its exceptionally high nutrient content. *Spirulina* is rich in various essential nutrients. *Spirulina* is renowned for its high protein content, making it an excellent source of plant-based protein. This protein is also considered complete as it contains all the essential amino acids. *Spirulina* contains a range of vitamins, including vitamin A, vitamin K, various B vitamins (such as B1, B2, B3, B6, and B12), and vitamin E. These vitamins play essential roles in various bodily functions, including immune function, energy metabolism, and vision. *Spirulina* contains essential fatty acids, such as omega-3 and omega-6 fatty acids, which are important for cardiovascular health, brain function, and inflammation regulation. In addition to being a rich source of protein, *Spirulina* contains a variety of amino acids, including both essential and non-essential amino acids. These amino acids are the building blocks of proteins and play crucial roles in various physiological processes. *Spirulina* is rich in minerals like iron, magnesium, potassium, calcium, and zinc. These minerals are essential for maintaining bone health, supporting immune function, and regulating various biochemical reactions in the body. *Spirulina* contains a range of phytonutrients, including chlorophyll, phycocyanin, and beta-carotene, which possess antioxidant and anti-inflammatory properties. These phytonutrients contribute to *Spirulina*'s potential health benefits and make it a valuable addition to the diet. *Spirulina* can indeed serve as an alternative or complementary source of nutrients to conventional vitamin supplements. Incorporating *Spirulina* into the diet can help enhance overall nutritional intake and support health and well-being. Additionally, its sustainable production makes it an environmentally friendly option for meeting nutritional needs.

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REFERENCES

1. UNICEF. Undernutrition. [On line] 2006 May [cited 2011 Dec 9]; Available at: URL: http://www.unicef.org/progressforchildren/2006n4/index_undernutrition.html
2. Simpore J, Zongo F, Kabore F, et al. Nutrition rehabilitation of HIV-infected and HIV-negative undernourished children utilizing *Spirulina*. Ann Nutr Metab 2005; 49: 373-380.
3. Belay A, Kato T, Ota Y. *Spirulina* (Arthospira): potential application as an animal feed supplement. J App Phycol 1996; 8: 303-11.
4. Ambrosi MA, Reinhr CO, Bertolin TE, et al. Health properties of *Spirulina* spp. Rev Cienc Farma Basica Apl 2008; 29: 109-17.
5. Hasler CM. Functional foods: the Western perspective. Nutr Rev 1996; 54(11): S6-10.
6. Chamorro G, Salazar M, Favila L, et al. Farmacología y toxicología del alga *Spirulina*. Rev Inv Clín 1996; 48: 389-99.
7. Salazar M, Martínez E, Madrigal E, et al. Subchronic toxicity study in mice fed *Spirulina* maxima. J Ethnopharmacol 1998;62: 235-241.
8. Jongkon P, Siripen T, Richard D L 2008 Maejo International Journal of Science and Technology 2(2) 159-171
9. Jung F, Krüger-Genge A, Waldeck P, Küpper J H 2019 Journal of Cellular Biotechnology 5 43-54 DOI 10.3233/JCB-189012
10. Tokusoglu Ö, üUnal M K 2006 Journal of Food Science 86(4): 1144 -1148