



Food Science and Applied Biotechnology

e-ISSN: 2603-3380

Journal home page: www.ijfsab.com
<https://doi.org/10.30721/fsab2019.v2.i2>



Research Article

Influence of *Spirulina Platensis* on the content of iron and zinc in wheat bread

Denka Y. Zlateva¹, Mimi P. Petrova^{1✉}, Dana A. Stefanova¹

¹Department of Commodities Science, Faculty of Economics, University of Economics - Varna, Bulgaria

Abstract

It has been found that nutrition does not provide the necessary quantities of some important trace elements, including iron and zinc. Iron is an essential element for all living organisms. It has a key role in the transport of oxygen and as a cofactor in many enzymes. Zinc is a component of more than 200 enzymes involved in the synthesis of proteins and DNA and is needed for the metabolism of growth factors. Enriching bread with mineral substances is a very good approach to overcoming the deficit and achieving adequate levels of intake. Different sources are used for this purpose, but there has been a growing interest in seaweed in recent years. The purpose of this study is to evaluate the effect of 2 and 4% *Spirulina* powder supplement on the iron and zinc content of bread made from wheat flour type 500. To determine the iron and zinc content, a highly sensitive ICP-AES method is used. Enrichment with 2% and 4% *Spirulina platensis* has been found to increase the amount of zinc in wheat bread respectively to $6.36 \pm 0.64 \text{ mg.kg}^{-1}$ and $6.77 \pm 0.68 \text{ mg.kg}^{-1}$, whereas its content in the control sample is $5.99 \pm 0.49 \text{ mg.kg}^{-1}$. The amount of iron in the enriched samples reached $15.9 \pm 1.59 \text{ mg.kg}^{-1}$ with the addition of 2% *Spirulina platensis* and $24.7 \pm 2.48 \text{ mg.kg}^{-1}$ with the addition of 4%, while in the control sample of bread was $7.22 \pm 0.58 \text{ mg.kg}^{-1}$. Practical applications: The addition of *Spirulina platensis* algae to wheat bread increases its biological value and helps to achieve iron and zinc content amounts close to the recommended daily intake levels.

Keywords: *Spirulina*, wheat flour, bread fortification, zinc, iron, ICP – AES method

✉Corresponding author: PhD candidate Mimi Plamenova Petrova; Department of Commodities Science, Faculty of Economics, University of Economics - Varna, 77 Knyaz Boris I blvd. BG-9002 Varna, Bulgaria, mobile: +359 882009696;
E-mail: m.petrova@ue-varna.bg

Article history:

Received 7 June 2018

Reviewed 4 August 2018

Accepted 13 December 2018

Available on-line 10 October 2019

<https://doi.org/10.30721/fsab2019.v2.i2.55>

© 2019 The Authors. UFT Academic publishing house, Plovdiv

Introduction

Food is one of the main determinants of human health, the duration and quality of life and the ability of the human body to withstand several adverse environmental impacts. However, for most of the foods normally consumed, there is an unbalanced relationship of essential nutrients or considered biologically active substances. Malnutrition is a major challenge worldwide and the number of chronically undernourished and malnourished people has been rising (FAO 2016). Almost 30% of the world population suffers from some form of malnutrition and of these, more than 2 billion people suffer from micronutrient deficiencies, of which 52% are pregnant women and 39% are children under five years of age (FAO 2016). Iron is an essential element for all living organisms. The recommended daily intake is 8 mg for men and 18 mg for women according to Ordinance 1 of January 22, 2018 on the physiological norms for nutrition of the population in our country. In connection with its essential role in the transport of oxygen and as a cofactor in many enzymes, iron plays an important role in the maintenance of the immune system. The digestibility of iron in the gastrointestinal tract depends on the form and solubility of the compound under which it is present in the ingested food (Gandy et al. 2012), increased by the presence of ascorbic acid and other organic acids (lactic acid, and others). Zinc is involved in all major biochemical metabolic processes in the human body. Well-known is the important role that it plays for the health and the wide range of biological functions it performs. In this regard, the European Food Safety Authority (EFSA) presents an official position on health claims and the importance of zinc for human health. The main role of zinc is related to the fact that it is a component of over 200 enzymes that are related to the synthesis of proteins and DNA (Prasad 2014), with hormone synthesis and metabolism of growth factors in child development (Salgueiro et al. 2002). The relationship between adequate zinc intake and maintenance of normal

hair and nail growth as well as skin health (Wolfgang and Sandstead 2006) is established. The recommended daily intake is 8 mg for women and 11 mg for men, in accordance with the Ordinance 1 of 22 January 2018 on the physiological standards of nutrition of the population. According to the World Health Organization (WHO), zinc deficiency ranks 11th among the 20 most important risk factors contributing to the burden of disease in the world and 5th among the 10 most important factors in developing countries, while iron deficiency ranks 6th. Zinc deficiency is responsible for many severe health complications, including impairments relating to physical growth, the immune system and learning abilities, as well as an increased risk of infections, DNA damage and cancer development (Gibson 2006). On the other hand, iron deficiency is the most common cause of anemia globally. Anemia affects around 1.6 billion people worldwide, with pre-school children and pregnant women at the greatest risk (McLean et al. 2009). High consumption of bread is a national characteristic of the nutrition in Bulgaria. That is why the enrichment of bread with minerals is a very good approach to overcoming the deficit and achieving adequate levels of intake. The outstanding nutritive value and high content of bioactive components of the algae is already well known as several studies were focused on the analysis of their important components which have beneficial effects on human health like minerals, proteins, lipids and vitamins (Vonshak 1997; Ajeesh et al. 2009). *Spirulina platensis* is a microscopic blue-green algae that grows intensely in alkaline waters and gets its name from its spiral shape. *Spirulina platensis* was declared by the United Nations World Food Conference of 1974 as the best food for the future and United Nations World Health Organization (WHO) stated that *Spirulina platensis* represents an interesting food for multiple reasons, for example, it is rich in iron and protein and is able to be administered to children without any risk (Geneva, Switzerland June 8th 1993). During the

sixtieth session of the United Nations General Assembly (Second Committee, Agenda item 52), IIMSAM (International institution for the use of microalgae *Spirulina* against malnutrition) initiated a revised draft resolution on the 'Use of *Spirulina* to combat hunger and malnutrition and help achieve sustainable development' (Habib et al. 2008). As a follow-up on this resolution, the United Nations Food and Agriculture Organization (FAO) prepared a draft position on *Spirulina* that was presented in 2008. Thus, microalgae can be an alternative to achieve food of high quality with a low environmental impact because they can be cultivated in non-cultivable lands. *Spirulina* may be used in foods because it is a source of lots of nutrients, including minerals (Belay 2002). According to Barakat and co-authors (Barakat et al. 2016) minerals content of dried *Spirulina platensis* algae (mg.100g⁻¹) is: Na (859 mg), K (1399 mg), Ca (689 mg), P (960 mg), Mg (400 mg) and Fe (122 mg) as average. These data are in harmony with others obtained by Tokusoglu and Ünal (2003) who found that minerals content in *Spirulina platensis* is as follows: Na (929.4 mg), K (1412.9 mg), Ca (826.3 mg), P (750.7 mg), Mg (388.9 mg) and Fe (95.37 mg) as average. More recent studies have attempted to improve the nutritional properties of food products by adding microalgae biomass. Fradique et al. incorporated microalgae biomass, such as from *Chlorella vulgaris* and *Spirulina maxima* which yielded products with better chemical composition without affecting baking quality. To offer gluten-free bread to consumers with celiac syndrome, authors developed products with rice flour to replace wheat flour and with 2.0 to 5.0% *Spirulina platensis* added (Fradique et al. 2010). Another study was proposed by Burcu et al. (2016). The authors found that adding 10% of *S. platensis* can enhance nutritional quality of bread without a negative impact on the shelf life. Calcium, magnesium and iron contents of bread with *S. platensis* were 721.2; 336.6 and 41.12 ppm, while conventional bread contained 261.7 ppm calcium, 196 ppm magnesium and 8.72 ppm iron. The purpose of this study is to evaluate the effect of 2 and 4% *Spirulina platensis* powder supplement on the iron and zinc content of bread made from wheat flour type 500.

Materials and Methods

Materials

In the course of the research the bread is made from wheat flour type 500 by a biphasic method. Initially, yeast dough is mixed with flour and water in a 1: 1 ratio. *Spirulina platensis* powder in a quantity of 2% or 4% of the flour mass is added in advance to the mixing water. The dough thus prepared matures at 33°C for 4 hours and then the bread dough is mixed by adding the remainder of the flour to the recipe and the salt (1.3 kg.100⁻¹ kg of flour). The bread dough matures for 50 min at 38°C. The bread is baked for 30-35 minutes at a temperature of 220°C (until the temperature in the bread crumb center reaches 96-98°C). For the purposes of the present study, the bread samples have been prepared and tested as follows: control sample - prepared only from wheat flour type 500, water, yeast and salt; as well as the enriched samples, in which, in addition to the mentioned raw materials, *Spirulina platensis* algae is included in the amount of 2% and 4% of the flour mass.

Method

To determine the content of iron and zinc a validated multistep analysis methodology was used using the ICP-AES method. Mineralization of the sample is performed according to BSS EN 13 805:2015 "Food products. Determination of trace elements. Digestion under pressure." It includes ashing in a microwave system and dissolving the ash residue in HNO₃. The resulting mineralase is injected into the plasma. The emission at 259.94 nm for Fe and 213.86 nm for Zn was measured. 3-5 consecutive measurements are made and the results averaged.

Results and Discussion

The results obtained in determining the iron and zinc content in the bread samples prepared with the addition of *Spirulina platensis* are presented in Figure 1. The data show that the inclusion of *Spirulina platensis* in the bread formulation leads to an increase in iron content. When enriched with 2% *Spirulina platensis*, the amount of iron in the bread is almost doubled, and the addition of 4% results in an increase in the amount by 3.4 times compared to

the control sample, reaching an amount of 24.7 mg.kg⁻¹. Minh (2014) also obtained similar results.

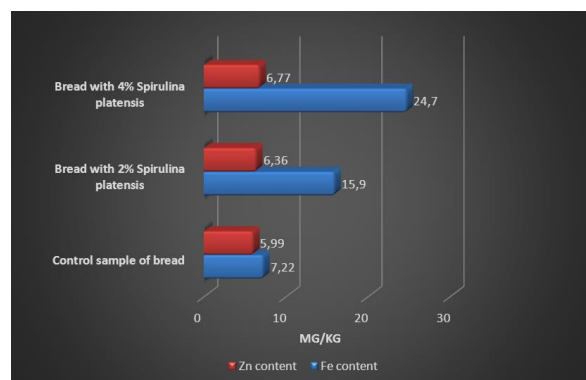


Figure 1. Content of Zinc and Iron in the analyzed samples (mg.kg⁻¹) determined using ICP-AES

He found that the addition of 3% *Spirulina platensis* increased the iron content to 27.0 mg.kg⁻¹, whereas the control contained 15.0 mg.kg⁻¹. In another research, Burcu Ak found that approximately 5 times more iron was detected in bread with 10% *Spirulina platensis* (Burcu et al., 2016). The differences in the quantitative data reported by the cited authors and those obtained by us are due to the differences in the composition of the algae used. Garcia-Casal et al. (2007, 2009) measured seasonal differences in the iron content of four seaweed species common to Venezuelan waters. Cabrita et al. (2016) found that iron contents varied, in some cases substantially, among macro algal species collected at the same sites and time, presumably linked to differences in metabolic requirements. It is of interest to compare the amounts thus obtained with the recommended daily intake of iron. Table 1 compares the levels of iron found in the bread samples tested and the recommended dietary intake levels, according to the Ordinance No. 1 of the Ministry of Health of January 22, 2018, on the physiological norms of nutrition of the population. For men aged 19-60 years, it is recommended intake of 8 mg of iron daily. Considering that the average daily bread consumption in Bulgaria is approximately 250 g, it can be concluded that only

22.56% of the recommended intake of iron are provided in the consumption of wheat bread. When enriched with 2% *Spirulina platensis*, the daily amount of bread consumed would provide almost half of the recommended iron intake, and the sample with 4% *Spirulina platensis* provides almost 80%. Women are given a higher recommended daily intake of 18 mg iron. From the study, it is clear that by enriching the bread with 4% *Spirulina platensis*, almost 35% of the recommended intake is provided, while the bread from wheat flour type 500 provides only 10%. From the results presented in Figure 1 it is clear that the zinc content in the control sample of bread is 5.99 ± 0.49 mg.kg⁻¹. Mohammed (2009) determines the content of macro- and microelements in white flour bread and establishes comparable Zn values (0.61 mg.100 g⁻¹). Ebuehi et al. (2007) investigate the mineral content of the most commonly consumed bread in Nigeria and establish zinc content that is of the same order as those obtained in our study (0.3-0.6 mg/100 g). Higher values were published by Demiröz et al. (2003) - 10 mg.100 g⁻¹. This may be due to the differences in soil-climatic conditions when growing wheat crops, as well as to the features of the technological processing of grain milling and flour production. By adding *Spirulina platensis*, the zinc content of wheat bread increases, albeit not as significantly as the iron content. This is quite normal, since *Spirulina platensis* algae have a much higher iron content than zinc - 49.50 ± 6.61 mg.100 g⁻¹ iron and 3.95 ± 0.25 mg.100 g⁻¹ zinc (Saharan and Jood, 2017). Incorporation of 2% *Spirulina platensis* into the bread formulation results in an increase in the amount of zinc by 0.37 mg.kg⁻¹ compared to the control sample, whereas with the addition of 4% of *Spirulina platensis*, the zinc content in the bread is increased by 13% relative to the control sample. Joshi et al. (2014) publish research data close to our results. They enriched corn flour biscuits with *Spirulina platensis* and found that the amount of zinc in biscuits reached levels of 6.66 mg.kg⁻¹. Table 2 compares the amount of zinc found in the control sample with the *Spirulina platensis* fortified breads and the recommended daily intake according to Ordinance No. 1 of 22 January 2018 on the physiological norms of nutrition of the population.

Table 1. Comparison between the iron content of the bread samples tested and the recommended daily intake

Samples tested	Iron content, mg/kg	Achieved levels compared to the recommended daily intake,%	
		For men aged 19-60	For women aged 19-60
Control sample bread	7.22 ± 0.58	22.56 %	10.02 %
Bread with 2% <i>Spirulina platensis</i> added	15.9 ± 1.59	49.69 %	22.08 %
Bread with 4% <i>Spirulina platensis</i> added	24.7 ± 2.48	77.18 %	34.31 %

Table 2. Comparison between the zinc content of the bread samples tested and the recommended daily intake

Samples tested	Zinc content, mg/kg	Achieved levels compared to the recommended daily intake,%	
		For men aged 19-60	For women aged 19-60
Control sample bread	5.99 ± 0.49	13.64 %	18.75 %
Bread with 2% <i>Spirulina platensis</i> added	6.36 ± 0.64	14.45 %	19.88 %
Bread with 4% <i>Spirulina platensis</i> added	6.77 ± 0.68	15.46 %	21.25 %

Men are scheduled to have a daily intake of 11 mg and for women 8 mg zinc. It is clear from Table 2 that bread with 4% *Spirulina platensis* provides over 15% of the recommended daily intake for men and over 21% for women, whereas in the control sample these values are lower (13.61 - 18.75%).

Conclusions

Based on these studies *Spirulina platensis* powder is found to be an alternative source that can be successfully used to increase the iron and zinc content in bread made from wheat flour type 500. The inclusion of algae in the bread formulation can

be successfully applied to overcome the deficiency of some trace elements, respectively - to increase its nutritional and biological value. Consumption of the enriched bread reveals an opportunity to overcome iron and zinc deficiency in nutrition and to reach levels much closer to the recommended daily intake, especially considering that bread is a product of high and daily consumption.

Acknowledgements

Authors would like to thank to the Ministry of Education and Science of Bulgaria about the subsidy by Regulation № 3 from 27.11.2015 – in accordance with the Ordinance on the Terms and

Procedure for the Evaluation, Planning, Allocation and Expenses of the State Budget Funds for the Financing of the Inherent Research Activities of the State Higher Education Institutions. The acknowledgements are also to the academic management of University of Economics – Varna for the allocations in project NPI-16/2018 “Innovative approaches to increase the quality and biological value of foods”.

References

- Ajeesh M, Bohra CPN, Gupta N, Rajasekaran C. *Spirulina* as 'functional food'. *New Biotechnol*, 2009, 25:S285, <https://doi.org/10.1016/j.nbt.2009.06.644>
- Barakat E., Nemaat M., El-Kewaisny, Salama A. Chemical and Nutritional Evaluation of Fortified Biscuits with Dried *Spirulina Algae*. *Food and Dairy Sci.*, Mansoura Univ ,2016, 7 (3): 167–77. www.journalijar.com/uploads/387_IJAR-18106.pdf
- Belay A. The Potential Application of *Spirulina* (*Arthrospira*) as a Nutritional and Therapeutic Supplement in Health Management. *The Journal of the American Nutraceutical Association*, 2002, 5(2), pp. 27-48, <http://www.macoc.fr/resources/The+Journal+of+the+American+Nutraceutical+Association+vol+5+spring+2002+Sp+inline.pdf>
- Burcu A., Avşaroğlu E., Işık O., Özyurt G., Kafkas E., Etyemez M., Uslu L. Nutritional and Physicochemical Characteristics of Bread Enriched with Microalgae *Spirulina Platensis*. *Journal of Engineering Research and Application*, 2016, 6(124): 2248–962230. http://www.ijera.com/papers/Vol6_issue12/Part-4/E612043038.pdf
- Cabrita A, Maia, M. R. G, Oliveira, H. M, Sousa-Pinto, I., Almeida, A. A, Pinto, E., & Fonseca, A. J. M. Tracing seaweeds as mineral sources for farm-animals. *Journal of applied phycology*, 2016, 28(5), 3135-3150. <https://doi.org/10.1007/s10811-016-0839-y>
- Demiröz B., Saldamlı I., Gürsela B., Uçak A., Çetinyokuş F., Yüzbaşı N. Determination of Some Metals Which Are Important for Food Quality Control in Bread. *Journal of Cereal Science*, 2003, 37(2): 171–77. <https://doi.org/10.1006/jcrs.2002.0491>
- Ebuehi, O., Owolabi O., Ikanone C., Amabibi I., Ajekwu A. Organoleptic, Minerals and Vitamins' Evaluation of Some Nigerian Breads, *Nigerian Food Journal*, 2007, 25(2). <http://dx.doi.org/10.4314/nifo.v25i2.50846>
- Fradique M, Batista AP, Nunes MC, Gouveia L, Bandarra NM, Raymundo A. Incorporation of *Chlorella Vulgaris* and *Spirulina Maxima* Biomass in Pasta Products. Part 1: Preparation and Evaluation. *Journal of the Science of Food and Agriculture*, 2010, 90(10): 1656–64. <https://doi.org/10.1002/jsfa.3999>
- Gandy J., Madden A., Holdsworth M. Oxford Handbook of Nutrition and Dietetics. Oxford University Press, 2012, Print ISBN-13: 9780199585823 <https://doi.org/10.1093/med/9780199585823.001.0001>
- García-Casal M, Pereira A, Leets I, Ramírez J, Quiroga M. High Iron Content and Bioavailability in Humans from Four Species of Marine Algae. *The Journal of Nutrition*, 2007, 137(12): 2691–95. <https://doi.org/10.1093/jn/137.12.2691>
- García-Casal M., Ramírez J., Leets I., Pereira A., Quiroga M. 2009. Antioxidant Capacity, Polyphenol Content and Iron Bioavailability from Algae (*Ulva Sp.*, *Sargassum Sp.* and *Porphyra Sp.*) in Human Subjects. *British Journal of Nutrition*, 2009, 101(01): <https://doi.org/10.1017/S0007114508994757>
- Gibson R. Zinc: The Missing Link in Combating Micronutrient Malnutrition in Developing Countries. The Proceedings of the Nutrition Society, 2006, 65(1): 51–60. <https://doi.org/10.1079/PNS2005474>
- Habib M., Parvin M., Huntington T., Hasan M. A review on culture, production and use of *spirulina* as food for humans and feeds for domestic animals. FAO Fisheries and Aquaculture Circular. No. 1034. Rome, FAO. 2008. 33p. ISBNs 9789251061060, www.fao.org/3/a-i0424e.pdf
- Joshi, S., Bera M. and Panesar P. Extrusion Cooking of Maize/*Spirulina* Mixture. *Journal of Food Processing and Preservation*, 2014, 38: 655-664. <https://doi.org/10.1111/jfpp.12015>
- McLean E., Cogswell M., Egli I., Wojdyla D. Worldwide Prevalence of Anaemia, WHO Vitamin and Mineral Nutrition Information System, 1993–2005. *Public Health Nutrition*, 2009, 12(04): 444. 3), <https://doi.org/10.1017/S1368980008002401>
- Minh N. Effect of *Saccharomyces Cerevisiae*, *Spirulina* and Preservative Supplementation to Sweet Bread Quality in Bakery. *International journal of multidisciplinary research and development*, 2014, 1(4): 36–44. <http://www.allsubjectjournal.com/archives/2014/vol1/issue4/70>
- Mohammed S., Mussali A., Mohsen A., Gahri A. Nutritive Value of Commonly Consumed Bread in Yemen. *E-Journal of Chemistry*, 2009, vol. 6, no. 2, pp. 437-444. <https://doi.org/10.1155/2009/975960>
- Prasad S. Impact of the Discovery of Human Zinc Deficiency on Health. *Journal of trace elements in medicine and biology: organ of the Society for Minerals and Trace Elements (GMS)*, 2014, 28(4): 357–63, <https://doi.org/10.1016/j.jtemb.2014.09.002>
- Saharan V. and Jood S. Nutritional Composition of *Spirulina Platensis* Powder and its Acceptability in Food Products. *International Journal of Advanced Research*, 2017, 5(6): 2295–2300, <http://dx.doi.org/10.21474/IJAR01/4671>
- Salgueiro M., Zubillaga M., Lysionek A., Caro R., Weill R., Boccio J. The Role of Zinc in the Growth and Development of Children. *Nutrition* (Burbank, Los Angeles County, Calif.) 2002, 18(6): 510–19. [https://doi.org/10.1016/S0899-9007\(01\)00812-7](https://doi.org/10.1016/S0899-9007(01)00812-7)

- Tokuşoglu, O., and Unal M. Biomass Nutrient Profiles of Three Microalgae: *Spirulina Platensis*, *Chlorella Vulgaris*, and *Isochrysis Galbana*. *Journal of Food Science*, 2003, 68(4): 1144–48.
<https://doi.org/10.1111/j.1365-2621.2003.tb09615.x>
- Vonshak A. *Spirulina Platensis (Arthrospira)*: Physiology, Cell-Biology, and Biotechnology. CRC Press, 1997, p.223, ISBN 9781482272970
www.bashanfoundation.org/contributions/Vonshak-A/1997-.Vonshak-S.pdf
- Wolfgang M. and Sandstead H. Zinc Requirements and the Risks and Benefits of Zinc Supplementation. *Journal of Trace Elements in Medicine and Biology* 20(1): 3–18.
<https://doi.org/10.1016/j.jtemb.2006.01.006>