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The effect of incorporating spirulina in Mozzarella cheese as a functional food

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Abstract

Microalgae (including spirulina) contain valuable bioactive compounds that act as potential antimicrobial agents.The aim of the current study was to prepare a functional Mozzarella cheese from cow's milk with incorporated spirulina and to analyze its effects on the microflora and on the sensory and physico-chemical properties.The spirulina presence in the Mozzarella cheese influenced significantly its color and taste, and also generated a higher moisture content. Considering the microbiological aspects, after the determination of the total number of germs and coliforms, the antibacterial effect was observed as a result of the spirulina addition; also the staphylococci, yeasts and molds presence was limited, and the inhibitory effect was accentuated by the addition of salt. *Spirulina platensis* has been shown to have antimicrobial activity against many pathogens (bacteria and fungi) and at the same time it increases the nutritional value of food.

Keywords: spirulina, Mozzarella cheese, functional food

1. Introduction

Mozzarella cheese belongs to the category of scalded cheese, originating in Southern Italy and obtained from buffalo or cow's milk. This fine type of cheese, with a subtle flavor can be used in a variety of recipes such as pizza, pasta, salads, including meat, seafood and vegetables.In recent years, food scientists have tried to fortify dairy products [20] by inoculating various compounds of plant origin to increase the sensory benefits and nutritional value of the products. Some flavoring and savoring substances have been added to Mozzarella to differentiate the product and make it more attractive [2]. Thus, spirulina incorporation in the dairy products has become a fairly common practice due to its sensory effects, but also to the functional properties that gives to food.

Spirulina offers important properties through its antimicrobial, antioxidant, anticancer and antihypertensive activity, strengthens the immune system, along with other pharmacological effects being included in the category of superfoods [1, 19]. Microalgae are known to be rich in proteins, amino acids, vitamins and various minerals, as well as polysaccharides, sterols and polyunsaturated fatty acids as bioactive compounds and pigments as coloring agents with important functional characteristics [5, 9]. Spirulina derived bioactive peptide fractions can be applied as nutraceutical ingredients in food and pharmaceuticals [13].

Preservatives are commonly used in food to inhibit microbial growth, and the algae extracts have been shown to be successful as antimicrobial agents against foodborne pathogens [12]. The main objective of the current study was to produce a functional Mozzarella cheese type by incorporating spirulina and to study its effect on the microflora and on the sensory and physical-chemical properties of the product obtained.

2. Materials and Method

2.1 Sample preparation

In order to carry out this research, 4 types of Mozzarella cheese were prepared from cow's milk, simple and with addition of spirulina powder. The milk was heated at 33-35^{0}C, and then the liquid curd was added, which must be dispersed evenly. After a while, the curd is formed, and it is cut into cubes with a side of 4-5 cm. Then the temperature is raised at 42 ° C and the curd is kept at that temperature for one hour; during this time the whey is removed.

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Then the whey is drained, and the curd is formed andcut into small pieces. After the curd is cut, the scalding process follows, and the pieces of curd are kneaded to form a soft paste. After the dough is formed, it is shaped into pieces of cheese, then placed into a bowl with cold water.

After the paste hardened, some of the samples were placed in a container with unsalted whey and others in whey with added salt. The same procedure was followed for two other samples of Mozzarella cheese in which 0.5% spirulina powder was incorporated before the milk coagulation.

When the small balls of Mozarella cheese with the addition of spirulina were shaped, part of them were introduced into unsalted whey and another part into salted whey. Thus, for that analysis, four samples of Mozzarella cheese were prepared: a simple control sample, another simple with added salt, respectively a functional one with spirulina incorporated without salt, and another one with spirulina and salt. The steps taken during cheeses preparation can be seen in Figure 1.

Figure 1. The stages of preparing functional mozzarella cheese

2.2 Sensory and physical-chemical evaluation

The descriptive sensory analysis was performed by tasters familiar with scoring methods and sensory language [7]. The sensory analysis was performed on each sample by 10 trained tasters, aged between 20 and 25 years.

The determination of the dry matter was performed by the oven drying method, through the difference being established the moisture content too.

2.3 Microbiological examination

The total number of germs was determined by Plate Count Agar (Standard Methods Agar), according to SR ISO 4833-1:12013. After incubation for 48 hours, the colonies developed under aerobic conditions were counted. The microbiological analysis for the determination of the number of yeasts and molds that can cause alteration of Mozzarella cheese was performed according to a standardized method [STAS ISO 7954-2001] using Sabouraud Agar with Chloramphenicol as culture medium, incubated at 25°C for 5 days. Coliform counts were performed according to the national standard for the enumeration, method SR EN ISO 4831: 2009. The enumeration of coagulase-positive staphylococci was performed according to SR EN ISO 6888-1:2002; it was made using Baird-Parker agar base with egg yolk tellurite, incubated at 37°C for 48 h. Analyses were performed in triplicate, and the mean values obtained were logarithmic.

3. Results and Discussion

3.1. Sensory and physical-chemical evaluation

Actually, the sensory properties of Mozzarella cheese can be influenced by several factors, including the feeding system of the animal from which the raw material originates [6]. The addition of spirulina can greatly influence the sensory properties of food, because like most microalgae, it has an unwanted odor. In fact, scientific researchers have investigated several methods of odor reduction that have been proved to be effective [3], and spirulina appropriate concentrations do not affect the sensory quality of products.

The results of this present study considering the average scores obtained for each sample of Mozzarella cheese in terms of appearance, taste, color, consistency and smell are shown in Figure 2.

Mozzarella cheese with spirulina and no added salt was the most appreciated by consumers in terms of color, appearance and taste, compared to simple samples without microalgae powder.

In terms of smell, simple and salty Mozzarella was more appreciated, as well as its consistency. As described by Bosnea *et. al.*, 2021 [5] cheeses with 0.25% and 0.5% incorporated spirulina were largely preferred by tasters because they had a less intense effect.

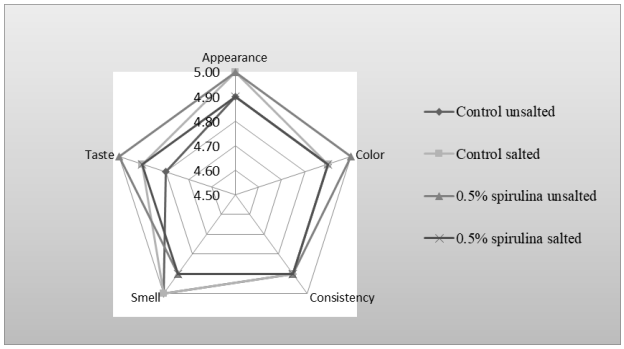

Figure 2. Diagram of the organoleptic analysis of different Mozzarella cheese

The moisture content varies depending on the ingredients added to the cheese, so Mozzarella with incorporated spirulina and salt added has the highest percentage, compared to the control samples without spirulina (Figure 3). The effect of *Spirulina platensis* on the humidity increasing of cheeses was also described by Tohamy *et. al.*, 2019 [21], who recorded values between 50-55%. The best consistency was registered by the simple and salty Mozzarella cheese with a dry matter content of over 55%. Increasing the dry matter content in all samples would be possible with the extension of the storage time, as there would be a decrease in humidity through the evaporation process after the maturation phase [14].

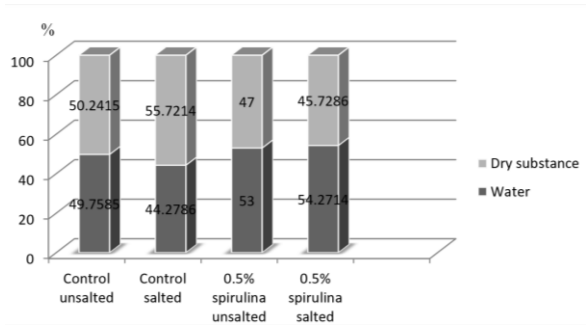

Figure 3. Results of moisture and dry matter content of Mozzarella samples

The evaluation of the microbiological quality and the safety of Mozzarella cheese, both simple and with incorporated spirulina, was performed by evaluating four parameters: total bacteria count, number of yeasts and molds, probable number of coliforms and the presence of staphylococci (CFU/g).

The analyses carried out report the presence of pathogenic and contaminating bacteria, and the logarithmic average values for the four types of Mozzarella (Table 1) provides indications on the microbiological profile of the product.

Table 1. Microbiological profile of Mozzarella cheese as a functional food

Mozzarella sample	Total bacteria count	Molds and Yeasts	*Coliforms*	*Staphylococci*
	Log10 cfu/g			
Control unsalted	4.32	2.81	1.48	0.3
Control salted	4.13	1.85	1.48	0
0.5% spirulina unsalted	4.09	2.48	1.04	0
0.5% spirulina salted	4.01	1.49	1.04	0

Thus, in the unsalted sample the number of germs decreases from 4.32 CFU/g to 4.09 CFU/g with the incorporation of spirulina in the product, and in the salty control sample from 4.13 CFU/g, the total bacteria count decreases to 4.01 CFU/g. The number of bacteria decreases in the samples of Mozzarella with spirulina powder addition and salt, that strengthens the inhibitory effect given by the seaweed added in the composition of the cheeses. Research by El-Fattah Elshouny *et al.*, 2017 [10] has shown that *Spirulina platensis* appears to be the most effective substance against the many studied pathogens.

The addition of spirulina has an inhibitory effect on staphylococcus that multiplies in cheese, thus, few colonies are identified in simple Mozzarella cheese. *Spirulina platensis* can inhibit the growth and development of Gram-positive bacteria including *Staphylococcus aureus* but also of the Gram-negative bacteria [4, 11]. In order to limit coliforms in the product, more attention must be paid to the degree of hygiene during its processing, but the reduction of the probable number of coliforms from 1.48 CFU/g to 1.04 CFU/ga was favored by the addition of spirulina, even if the processing was carried out under the same conditions. In the case of testing the degree of contamination with yeasts and molds, the efficiency of preservation was boosted with the addition of salt. However, there can be concluded that spirulina powder accentuates the antifungal effect by observing a reduction of yeast and molds colonies from 1.85 CFU/g in the simple salty sample to 1.49 CFU/g in the sample with the addition of spirulina and salt.

The inhibitory activity of spirulina extracts on fungal pathogens has been demonstrated against *Rhodotorula, Candida, Penicillium, Aspergillus* [8, 22] by diffusimetric methods. All these researches have contributed to the acceptance of microalgae as elements for fortifying dairy products.

4. Conclusion

Mozzarella cheese made from cow's milk has a local origin, and the addition of spirulina makes it an appreciable functional product.

The microbiological aspects such as the antibacterial and antifungal effects were observed due to the addition of spirulina in fresh Mozzarella, the inhibitory effect being accentuated by the addition of salt.

Spirulina platensis has been shown to have excellent nutritional, functional and healthy properties for its use as a nutraceutical food.

Compliance with Ethics Requirements. Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human or animal subjects (if exist) respect the specific regulation and standards.

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