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Application of Plant Ingredients for Improving Sustainability of Fresh Pasta

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Abstract: Pasta is a low-cost and easy-to-prepare food product. By using fresh pasta, the drying process is omitted, which represents significant energy and financial savings, but the durability of such pasta is very limited. The addition of plant materials (parsley, oregano, thyme, cinnamon, nettle, spinach, and carob) to the dough affects the microbiological stability and thus the durability of the fresh pasta, which can then be stored longer in the refrigerator without the need to freeze the product. With significant energy savings due to the omission of the drying and freezing processes, extending the shelf life of the pasta will contribute to the reduction of food waste and thus contribute to a more sustainable production system. The aim of this work was to examine the possibility of producing fresh pasta with the addition of plant materials from the perspectives of technological and sensory quality, as well as microbiological composition. The incorporation of plant materials (10 g/100 g) decreased water absorption and had no effect on optimal cooking time. Thyme-enriched pasta had significantly lower ($p < 0.05$) cooking loss (3.34%) than the control sample (4.12%). In cinnamon-, spinach-, and parsley-enriched pasta, an appealing colour and pleasant smell and taste were achieved. Cinnamon and carob proved to have the most favourable effect on the microbiological quality (reduction in mesophilic, *Enterobacteriaceae*, and yeast counts), followed by spinach (reduction in mould count). For cinnamon-enriched pasta, the best technological (optimal cooking time of 3.23 min; cooking loss of 4.41%; firmness of 531.94 g) and sensory and microbiological quality were achieved; thus, cinnamon proved to be the preferred natural preservative for the production of enriched pasta.

Keywords: cooking properties; fresh pasta; microbiological quality; plant ingredients; sensory evaluation

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1. Introduction

Growing consumers' awareness regarding diseases caused by inappropriate nutrition is a driving force in the search for enriched food products. The most suitable foods for enrichment are those that are popular worldwide, are consumed on a daily basis, and whose sensory properties are acceptable to consumers regardless of age. In this regard, the food industry is facing the challenge of how to produce foods with diverse functionality while maintaining the consumer acceptability of such foods [1]. The sensory properties of new products must meet consumer expectations; therefore, the improvement of nutritional value using already familiar ingredients represents a straight-forward strategy.

Pasta is a staple food rich in complex carbohydrates, a moderate source of protein and vitamins, low in sodium and fat, and free of cholesterol [2,3]. In addition to its low cost and ease of preparation [4], pasta is considered a valuable energy source in human nutrition [2] and food with a low glycemic index [5,6]. As such, pasta is a product very suitable for enrichment with various ingredients with a high content of dietary fibres, antioxidants, vitamins, and minerals. However, the durability of fresh pasta is very limited. Although significant energy savings are achieved during the production of fresh pasta, because there is no drying process, this energy savings may not be a complete alternative to drying, as fresh pasta must be stored in the refrigerator and dry pasta has a much longer shelf life at ambient temperature. The shelf life of fresh pasta strictly depends on the microorganisms that grow during storage and can be significantly extended by using preservatives [7]. The FDA prescribes the use of chemical preservatives in the production of fresh pasta [8]. However, nowadays society desires fewer synthetic food additives, so new methods for safe food production are sought. A possible alternative is the use of natural active ingredients such as different parts of plants, bioprotective cultures, and essential oils, which contribute towards sustainability [9,10]. For this purpose, many plant species (herbs, spices, fruits, and vegetables) were examined. Parsley, oregano, thyme, and cinnamon are highly valued worldwide as culinary ingredients and are characterised by the presence of bioactive compounds responsible for their additional health benefits. Parsley is rich in bioactive compounds exhibiting an antioxidant, hepatoprotective, brain-protective, antidiabetic, analgesic, gastroprotective, laxative, diuretic, antibacterial, and antifungal effect [11]. Bioactive compounds present in oregano, thyme, and cinnamon are effective antioxidants and antimicrobial agents [12–14]. Herbs and vegetables such as nettle, spinach, and carob are natural sources of phenolic compounds, carotenoids, minerals, vitamins, and dietary fibre [15–17]. However, besides the numerous benefits and high potential of plant-based ingredients, certain negative consequences and risks must be considered when it comes to their incorporation in pasta.

The improvement of the nutritional and functional properties of pasta with non-conventional ingredients is often accompanied by a deterioration in its technological quality [18–20]. The main disadvantages of enriching pasta with herbs and vegetables are the weakening of the gluten network and the impairment of starch gelatinization [4]. In order to limit or eliminate such disadvantages while maintaining already established quality standards for pasta [21], formulations with novel ingredients must be optimized. Furthermore, when the goal is the enrichment of pasta with plant ingredients, it is worth considering the production of fresh pasta because the loss of some bioactive compounds was reported as an effect of pasta drying [22]. On the other hand, fresh pasta is susceptible to rapid spoilage due to its high water content, which could lead to waste generation or even poisoning [23]. In order to prolong the shelf-life of fresh pasta, preservation at low temperatures (freezing) is often applied. However, this step increases investment costs, which is inconvenient, especially for small- and medium-scale producers. Moreover, even though many plant ingredients have proven antimicrobial activity, spices and herbs, particularly, possess high microbial contamination [24,25]. The incorporation of such raw materials into a product with a short shelf life represents a major challenge in terms of food safety.

Therefore, the aim of this study was to determine the possibility of producing sensory-acceptable-quality fresh pasta enriched with parsley, oregano, thyme, cinnamon, nettle, spinach, and carob, which would be in compliance with food safety requirements. Another aim of this research was the utilisation of the microbial potential of the mentioned plant ingredients and the evaluation of their effectiveness in the preservation of fresh pasta quality while stored in refrigerating conditions. The quality of the obtained plant-enriched fresh pasta was investigated on the basis of its pasta cooking properties, texture, sensory, and microbiological quality assessments.

2. Materials and Methods

2.1. Raw Materials

Raw materials used for the preparation of pasta were: wheat flour—farina (13.4% moisture, 11.8% protein); parsley; oregano; thyme; cinnamon; nettle; spinach in dry powdered form; and carob pod flour purchased at the local market in Novi Sad, Serbia.

2.2. Microbiological Analysis of Plant Ingredients

Microbiological quality of plant ingredients was assessed based on the determination of: (1) mesophilic bacteria [26] on plate count agar (Biolife, Milan, Italy) incubated for 3 days at 30 °C; (2) *Enterobacteriaceae* [27] on violet red bile glucose (Biolife, Milan, Italy) incubated for 1 day at 37 °C; (3) yeast and moulds [28] on dichloran rose-bengal chloramphenicol agar (Biolife, Milan, Italy) incubated for 5 days at 25 °C; (4) *Staphylococcus aureus* [29] on Baird–Parker agar (Biolife, Milano, Italy) incubated for 2 days at 37 °C; and (5) *Bacillus cereus* [30] on *Bacillus cereus* agar (Biolife, Milan, Italy) incubated for 1 day at 30 °C. The results of mesophilic bacteria, *Enterobacteriaceae*, yeast, and mould were counted and presented as log CFU/g, and the results of *S. aureus* and *B. cereus* were presented as positive (when typical growth has been observed on selective agar plates) or negative.

2.3. Pasta Preparation

Enriched pasta samples were prepared by replacing farina with 10 g/100 g parsley, oregano, thyme, cinnamon, nettle, spinach powder, or carob flour, while the control pasta was prepared from wheat flour only. The quantity of plant ingredients was chosen based on the recipes of commercially available pasta at the market in Serbia. In order to obtain optimally developed homogeneous dough with a moisture content of 31.5%, the amount of water required for the hydration of the mixtures was calculated for each sample using Equation (1):

$$V = \frac{F\left(w_t - w_f\right)}{100 - w_t} \tag{1}$$

where V represents water amount (kg), F is quantity of farina (kg), w_t is dough moisture content (%), and w_f is farina moisture content (%).

The pasta preparation was carried out in a professional pasta machine (La Parmigiana, Fidenza, Italy) equipped with an extruder and a bronze die for the tagliatelle form, under the following conditions: 15 min mixing; temperature 30 °C; 42 rpm auger extrusion speed. Obtained fresh pasta samples (about 2 mm thickness, 9 mm width, and 200 mm length) were packaged in plastic bags and refrigerated (4–5 °C) until analysis.

2.4. Pasta Cooking Properties

The evaluation of the pasta cooking properties (optimal cooking time, cooking loss, and water absorption) was performed according to AACC method 66-50.01 [31]. The optimum cooking time (OCT) was determined by cooking 100 g of fresh pasta sample in 1 L of boiling tap water until achieving the complete disappearance of the starchy central core. The cooking water was then first evaporated (80 °C) and then dried at 105 °C ± 2 °C until a constant weight was obtained to obtain the cooking loss (CL). Water absorption (WA) was calculated by subtracting the weight of cooked pasta after reaching the OCT from the weight of uncooked pasta [32].

2.5. Firmness of Cooked Pasta

The firmness of the cooked pasta was evaluated using a TA.HDplus Texture Analyser (Stable Micro Systems, Surrey, UK) equipped with a 5 kg load cell and Perspex blade according to the AACC 66-50.01 method [31]. Three tagliatelle pieces of each pasta sample were subjected to a firmness test 10 min after cooking under the following operating conditions: a test speed of 2 mm/s; distance from the platform of 0.1 mm; post-test speed of 5 mm/s. The pasta firmness was measured and calculated with the Texture Exponent software, Version 6.0.6.0 (Stable Micro Systems, Surrey, UK) from the graph as the height of the peak (in grammes), which is analogous to maximum cutting force [33,34].

2.6. Sensory Evaluation

The sensory attributes of pasta were evaluated by a trained panel of six members (ages 27–45, both genders), based on the grading scale of 1.0–5.0. The following quality attributes of pasta were evaluated: shape, colour, smell, taste, adhesiveness, chewiness, surface stickiness, and surface stickiness after 10 min. Each pasta sample was cooked according to the procedure described above (Section 2.4), drained, and cooled for 5 min at room temperature. Approximately 100 g of each pasta sample was placed on a white plate and presented to each assessor in a randomised order. Panellists were asked to try each sample and rate each attribute by circling the number on the scale. Between the tastings of pasta samples, the panellists were asked to rinse their mouths with water. The sensory evaluation was performed in separate booths under normal lighting conditions at room temperature (25 °C).

2.7. Pasta Microbiological Analysis

Pasta samples for microbiological testing were transported to the Laboratory for Food Microbiology, Biotechnical Faculty, University of Ljubljana, Slovenia. The samples were stored in the refrigerator, and sampling was carried out on days 1, 4, 8, and 12. The microbiological quality parameters of the pasta were determined by monitoring the mesophilic bacteria, *Enterobacteriaceae*, yeast, and moulds, *Staphylococcus aureus* and *Bacillus cereus*, using the same methods as for plant material, which are described in Section 2.2. The results of mesophilic bacteria, *Enterobacteriaceae*, yeast, and mould were counted and presented as a reduction in the number of microorganisms in relation to the control sample. The results of *S. aureus* and *B. cereus* were presented as positive (when typical growth has been observed on selective agar plates) or negative.

2.8. Statistical Analysis

Measurements of pasta physical characteristics were performed in triplicate, and the results were expressed as the mean value ± standard deviation. One-way analysis of variance (ANOVA) and Duncan's multiple range test were used (Statistica 13.5, TIBCO Software Inc., Palo Alto, CA, USA) to assess the significance of the differences between the mean values at a significant level of 0.05. Microbiological experiments were performed in duplicate. For each microbial parameter at each time point, plant-enriched pasta was compared to the control with an ANOVA and Dunett t-test at a significance level of 0.05 (SPSS V23, IBM Corporation, North Caste, NY, USA).

3. Results and Discussion

3.1. Pasta Cooking Properties

The results for the cooking properties of pasta are presented in Table 1. The OCT values for plant-enriched pasta were significantly lower than in previous reports [20,32,35–37], probably as a consequence of the use of wheat flour (farina) in pasta production and the smaller dimensions of the produced pasta samples. The substitution of wheat flour by 10 g/100 g of parsley, oregano, thyme, cinnamon, nettle, spinach, and carob led to no significant ($p > 0.05$) variation in the OCT.

Previous studies, however, report different findings. Boroski et al. [36] and Biernacka et al. [35] reported lower OCT values for pasta with 10% oregano leaf and pasta enriched with 1–5% carob fibre compared to the control, due to the presence of dietary fibre in plants, which causes disruption of gluten structure, leading to lower OCT [38]. Regardless of those facts, the expected decrease in OCT did not occur in our study. These values were not significantly different from the control sample ($p > 0.05$), because the cooking time of pasta made from farina wheat flour was certainly very short compared to pasta made from semolina, where the cooking time is about 15 min [39], so the influence of plant materials on the OCT of our pasta samples could not be so pronounced.

Table 1. Cooking properties and firmness of control and pasta enriched with 10 g/100 g parsley, oregano, thyme, cinnamon, nettle, spinach, and carob.

Plant-Enriched Pasta	Optimum Cooking Time (min)	Cooking Loss (%)	Firmness (g)
Control	4.4 ± 0.2^{a}	4.12 ± 0.16^{b}	813.41 ± 15.69^{cd}
Parsley	4.5 ± 0.3^{a}	4.76 ± 0.02^{e}	581.29 ± 18.98^{ab}
Oregano	4.3 ± 0.4^{a}	4.41 ± 0.04^{cd}	645.89 ± 12.74^{b}
Thyme	4.5 ± 0.2^{a}	3.34 ± 0.03^{a}	745.95 ± 11.57^{c}
Cinnamon	4.3 ± 0.3^{a}	4.41 ± 0.11^{cd}	531.94 ± 14.18^{a}
Nettle	4.3 ± 0.3^{a}	4.64 ± 0.03^{de}	524.18 ± 33.29^{a}
Spinach	4.4 ± 0.5^{a}	4.38 ± 0.03^{c}	846.57 ± 86.80^{d}
Carob	4.3 ± 0.4^{a}	5.35 ± 0.05^{f}	628.80 ± 32.81^{b}

Mean ± standard deviation, number of measurements = 3. $^{a–f}$ Within the same column, mean values with different superscript letters indicate a significant difference determined by the Duncan's test ($p < 0.05$).

The obtained CL values were similar to previous reports for pasta enriched with oregano and carrot leaf [36] and soluble dietary fibres [40], but lower compared to pasta enriched with olive pomace [32] and insoluble dietary fibres [41]. In all pasta samples, with the exception of thyme-enriched pasta, a significant increase ($p < 0.05$) in the CL compared to the control (Table 1) was found in accordance with previous reports for pasta enriched with plant and dietary fibre [32,36,42,43]. The disruption of the continuous gluten network by the incorporation of plant ingredients into the pasta leads to a reduction in the ability to retain amylose during cooking. Furthermore, higher CL values recorded in pasta samples enriched with parsley and nettle could be related to the potential leaching of phenolic compounds present in parsley [37] and nettle leaf [15]. The lowest CL value was recorded in thyme-enriched pasta, while the highest CL value was recorded in carob-enriched pasta. A possible explanation for the highest CL values in the carob-enriched pasta is that a high amount of dietary fibre in carob fruit disturbed the protein matrix. The obtained CL results for carob-enriched pasta were almost 1.7% higher than the results for pasta enriched with 1–5% carob fibre, as reported by Biernacka et al. [35]. It is important to note that all pasta samples had CL values below 8%, which is recognised as a technologically acceptable limit [44].

3.2. Firmness of Cooked Pasta

With the exception of spinach-enriched pasta, a decrease in the firmness of cooked pasta samples was observed compared to the control (Table 1). In his research, Wang [45] concluded that spinach-enriched pasta was generally firmer than the control sample, potentially due to its high protein and relatively low fibre nature. In our case, the addition of spinach did cause an increase in hardness, although this increase was not statistically significant ($p > 0.05$) compared to the control sample. The trend of decrease in firmness was previously reported for pasta with brewers' spent grain [46], inulin, and bran-semolina [40,42], and for pasta enriched with carob fibre [35] and apple pomace [47]. It is assumed that the presence of the dietary fibres in the plant ingredients interferes with the starch water absorption, which leads to a reduction in the pregelatinized starch and disturbs the gluten-starch matrix, resulting in a reduction in the mechanical strength and, thus, pasta firmness [20]. The highest value of pasta firmness was obtained for spinach-enriched pasta, followed by thyme and oregano-enriched pasta, and the lowest for nettle and cinnamon-enriched pasta. Obtained firmness values were comparable with literature results for inulin [40] and bran-semolina [41]-enriched pasta.

3.3. Sensory Evaluation

The results of the sensory evaluation of pasta samples are shown in Table 2. The higher scores for the evaluated attributes implied a higher acceptance of the pasta.

Table 2. Sensory evaluation of control pasta and pasta enriched with 10 g/100 g parsley, oregano, thyme, cinnamon, nettle, spinach, and carob.

Plant-Enriched Pasta.	Shape	Colour	Smell	Taste	Adhesivenes	Chewiness	Surface Stickness	Stickness after 10 min
Control	5.00 ± 0.00^{c}	5.00 ± 0.16^{a}	4.83 ± 0.41^{bd}	4.00 ± 0.00^{ac}	4.83 ± 0.41^{a}	5.00 ± 0.00^{a}	4.66 ± 0.52^{b}	3.16 ± 0.75^{ab}
Parsley	4.00 ± 0.00^{ab}	5.00 ± 0.00^{a}	4.00 ± 0.00^{a}	5.00 ± 0.00^{d}	4.83 ± 0.41^{a}	4.83 ± 0.41^{a}	3.00 ± 0.00^{c}	3.66 ± 0.52^{abc}
Oregano	4.33 ± 0.81^{abc}	4.83 ± 0.41^{a}	4.33 ± 0.52^{ac}	3.50 ± 0.84^{ab}	4.83 ± 0.41^{a}	4.66 ± 0.51^{a}	4.33 ± 0.82^{ab}	3.33 ± 0.51^{abc}
Thyme	3.83 ± 0.98^{a}	3.83 ± 0.75^{b}	4.00 ± 0.00^{a}	3.33 ± 1.03^{ab}	4.83 ± 0.41^{a}	4.00 ± 0.00^{b}	3.83 ± 0.75^{a}	3.00 ± 0.00^{a}
Cinnamon	4.00 ± 0.63^{ab}	5.00 ± 0.00^{a}	5.00 ± 0.00^{b}	5.00 ± 0.00^{d}	4.00 ± 0.00^{b}	4.50 ± 0.55^{ab}	3.66 ± 0.52^{a}	3.33 ± 0.52^{abc}
Nettle	4.16 ± 0.40^{ab}	5.00 ± 0.00^{a}	4.50 ± 0.53^{cd}	4.00 ± 0.00^{ac}	4.83 ± 0.41^{a}	4.66 ± 0.52^{a}	4.33 ± 0.52^{ab}	3.83 ± 0.41^{bc}
Spinach	4.66 ± 0.51^{bc}	5.00 ± 0.00^{a}	5.00 ± 0.00^{b}	4.66 ± 0.82^{cd}	4.83 ± 0.41^{a}	4.83 ± 0.41^{a}	3.83 ± 0.41^{a}	3.16 ± 0.75^{ab}
Carob	5.00 ± 0.00^{c}	5.00 ± 0.00^{a}	3.17 ± 0.41^{e}	3.17 ± 0.41^{b}	4.83 ± 0.41^{a}	4.00 ± 0.63^{b}	3.83 ± 0.41^{a}	4.00 ± 0.63^{c}

a–e Within the same column, mean values with different superscript letters indicate a significant difference determined by the Duncan's test ($p < 0.05$).

When assessing the shape of cooked pasta samples, the highest scores were obtained for control and carob-enriched pasta (5.0), followed by spinach-, oregano-, and nettle-enriched pasta (4.66, 4.33, 4.16, respectively, Table 2). The colour of the pasta is a very important characteristic that can greatly affect the acceptability of the product. The colour of the pasta was influenced by the incorporation of all plant ingredients (Figure 1), but panellists found that the colour of the plant-enriched pasta was very acceptable and attractive for all samples except for the thyme-enriched pasta, which was the only one that received a statistically significant lower score ($p < 0.05$) compared to the other samples (Table 2).

Figure 1. Appearance of cooked control an enriched pasta sample.

Cinnamon- and spinach-enriched pasta received the highest scores (5.0) for smell, considering that smell was recognisable and familiar as well as more intense, followed by control, nettle, and oregano-enriched pasta (4.83, 4.5, and 4.33, respectively, Table 2). Another very important feature of pasta is certainly the taste, and any addition can cause very significant changes. For example, the use of fish oil as an excellent source of omega-3 fatty acids in pasta is limited due to its unfavourable taste and high sensitivity to oxidation. In our study, the highest scores for taste were obtained in parsley and cinnamon-enriched pasta (5.0), while the taste of spinach-enriched, control, and nettle-enriched pasta was also very acceptable (4.44, 4.0, and 4.0, respectively, see Table 2). Carob-enriched pasta received the lowest scores for both smell and taste (3.16), probably due to the high tannin content in carob flour responsible for the bitter taste [48]. However, Sęczyk et al. [17] reported that substitution with carob flour at a lower level (1–5 g/100 g) could be used since a significant effect on pasta sensory attributes and acceptance was not observed. The deterioration in adhesiveness of pasta was minimal when plant ingredients were incorporated, but was more pronounced in cinnamon-enriched pasta (4.0, Table 2). The control pasta received the highest score for chewiness (5.0), followed by parsley- and spinach-enriched pasta (4.83), while the lowest scores were observed in thyme- and carob-enriched pasta (4.0).

The incorporation of plant ingredients increased the surface stickiness of pasta, which was more pronounced in parsley- and cinnamon-enriched pasta (3.0, 3.66, Table 2). The highest scores for chewiness and stickiness of control pasta were expected, as the sample value for cooking loss was the lowest (Table 1), and the addition of plant materials caused cooking loss to increase. The release of amylose during cooking, due to the weakened protein matrix with the addition of plant materials, leads to an increase in the stickiness of the pasta and a worse feeling of chewiness because the pasta falls apart during chewing. The highest surface stickiness after 10 min was found in thyme-enriched pasta (3.0), while carob-enriched pasta had the lowest surface stickiness after 10 min (4.0, Table 2). In general, pasta sensory attributes most affected by plant ingredient incorporation were colour (as can be seen in Figure 1), taste, and smell (the largest statistical difference between the scores is observed for these attributes in Table 2). Corresponding sensory attributes were positively influenced by the enrichment with parsley, cinnamon, and spinach, and these samples were characterised by an appealing colour and a pleasant and unique aroma. Conversely, thyme- and carob-enriched pasta received the lowest scores for the corresponding sensory parameters and consequently had lower acceptance. However, the presented results of the sensory evaluation showed that the inclusion of plant ingredients, especially parsley, cinnamon, and spinach, at a level of 10 g/100 g in pasta production can improve certain sensory attributes of plant-enriched pasta.

3.4. Microbiological Analysis of Pasta Samples and Plant Materials Used for Pasta Enrichment

The results of mesophilic bacteria, *Enterobacteriaceae*, yeast and mould, *S. aureus*, and *B. cereus* of plant materials used for pasta enrichment are presented in Table 3. The observed results are consistent with the study of Garbowska et al. [49], in which the authors determined higher contamination of several plants, especially herbs. According to the guidelines elaborated by the International Commission on Microbiological Specifications for Foods [50], the total bacterial count in spices below 10^4 CFU/g is defined as acceptable, the count of 10^4–10^6 CFU/g is defined as permissible, and the count exceeding 10^6 CFU/g is defined as unacceptable quality. So that all plant materials used in the production of our pasta samples were of acceptable and permissible quality. The results on the effect of plant ingredients on the fresh pasta microbiological quality after 1, 4, 8, and 12 days of storage are shown in Figure 2. Some of the ingredients improved the microbiological quality of pasta, while others deteriorated it. Spinach-, parsley-, and thyme-enriched pasta had a higher mesophilic bacterial count than the control sample already on the first day (Figure 2), which can be explained by the higher bacterial contamination of the mentioned plant ingredients themselves (Table 3). However, a decrease in the number of mesophilic bacteria in parsley- and thyme-enriched pasta compared to the control was observed after the fourth and eighth days of storage, which confirms the antimicrobial activity of parsley and thyme [51,52]. In the mesophilic bacterial count (Figure 2A), cinnamon and carob showed a very good effect in reducing bacterial growth until the last day of examination. Among the examined pasta samples, only spinach-enriched pasta had a higher count of mesophilic bacteria until day eight compared to the control, while the observed parameter in oregano-enriched pasta was similar to the control pasta throughout the sampling (Figure 2A).

Table 3. Microbiological quality of plant materials used for pasta enrichment.

Plant Materials	Mesophylic Bacteria (log CFU/g) *	*Enterobacteriaceae* (log CFU/g) *	Yeasts and Moulds (log CFU/g) *	*Staphylococcus aureus* **	*Bacillus cereus* **
Parsley	4.15 ± 0.05^{d}	2.00 ± 0.00^{a}	2.15 ± 0.05^{b}	+	+
Oregano	3.87 ± 0.09^{c}	2.37 ± 0.07^{b}	2.74 ± 0.08^{d}	+	+
Thyme	4.72 ± 0.03^{e}	−	2.52 ± 0.02^{c}	−	+
Cinnamon	2.00 ± 0.00^{a}	−	2.05 ± 0.01^{a}	−	+
Nettle	3.37 ± 0.07^{b}	2.72 ± 0.05^{c}	2.97 ± 0.07^{e}	−	+
Spinach	4.96 ± 0.08^{f}	2.15 ± 0.15^{a}	2.23 ± 0.05^{b}	−	−
Carob	2.00 ± 0.00^{a}	−	2.00 ± 0.00^{a}	+	+

* limit of detection 10 CFU/g. ** Results are expressed as positive (+) when typical growth has been observed on selective agar plates or negative (−). $^{a–f}$ Within the same column, mean values with different superscript letters indicate a significant difference determined by the Duncan's test ($p < 0.05$).

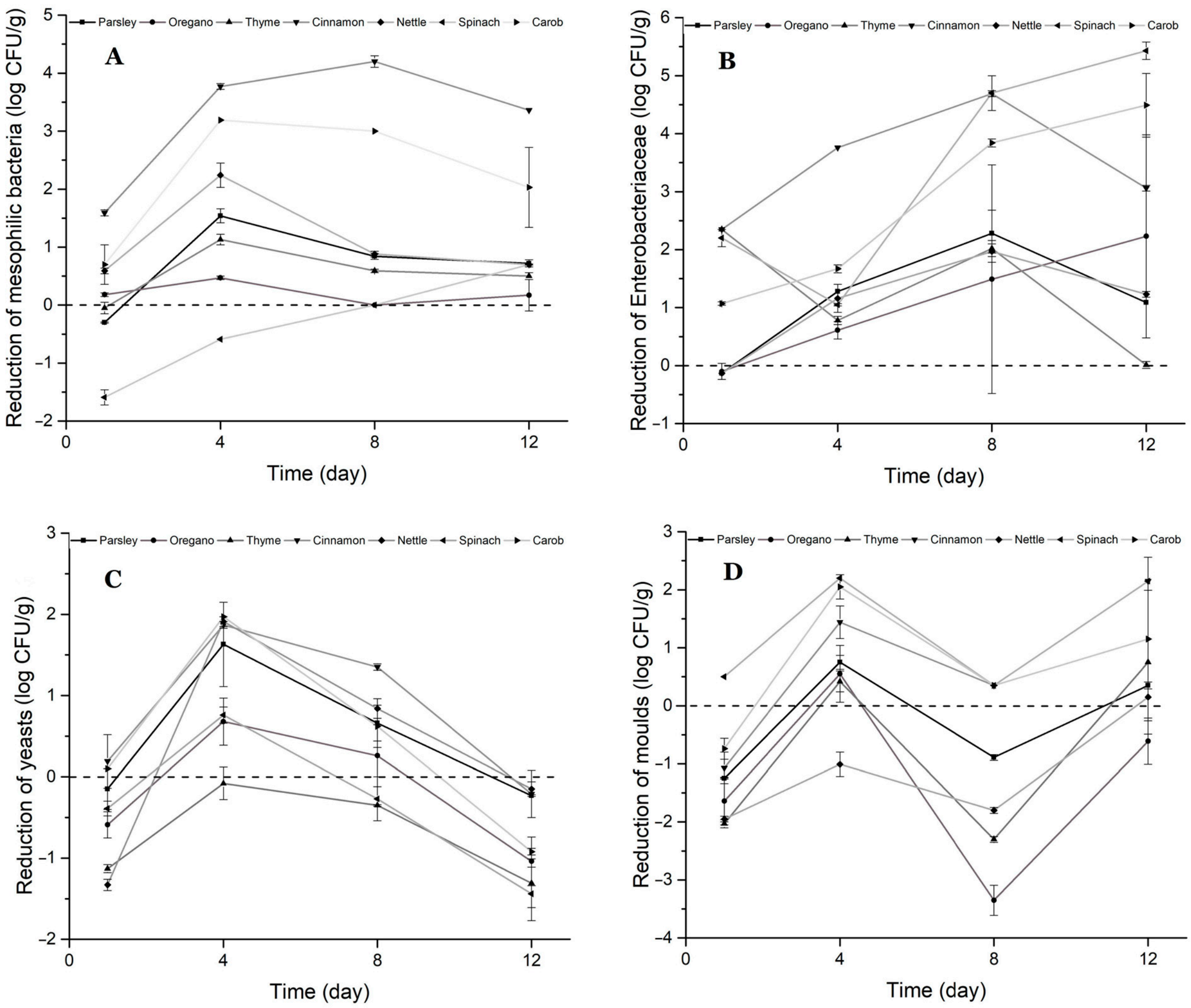

Figure 2. The effect of plant ingredients on the fresh pasta microbiological quality after 1, 4, 8, and 12 days of storage: reduction in the number of mesophilic bacteria (**A**), *Enterobacteriaceae* (**B**), yeast (**C**), and moulds (**D**) in relation to the control sample.

When observing the influence of plant ingredients on the number of *Enterobacteriaceae*, cinnamon and carob showed again the most pronounced positive effect (Figure 2B). It is worth mentioning that in the case of the mentioned plant ingredients (as well as thyme), no growth of *Enterobacteriaceae* was observed within the methods' limit of detection (Table 3). In other pasta samples, the number of *Enterobacteriaceae* was inconsistent throughout the sampling but mostly showed a reduction in the observed parameter during storage when compared to the control sample.

Furthermore, the use of cinnamon and carob also proved to have a positive effect on the growth inhibition of yeasts (Figure 2C). The use of parsley and nettle was shown to inhibit the growth of yeast from day four to day eight. However, there was a different pattern in the number of moulds, with spinach-enriched pasta proving to be the best sample (Figure 2D). On the first day of sampling, the mould load was higher for plant-enriched pasta compared to the control, probably due to the initial contamination of dry ingredients with moulds (Table 3) [53]. In addition, the presence of two potentially pathogenic bacteria, which can cause food poisoning, was monitored. Presumptive *S. aureus* could only be detected after 8 days of storage in control oregano- and carob-enriched pasta. Presumptive *B. cereus* was detected in all pasta samples except the control and spinach-enriched pasta. As with moulds, it is supposed that this is the consequence of raw material contamination with *Bacillus* and *Staphylococcus* spores [54]. All plant ingredients used in the present study are known for their in vitro antimicrobial activity against *Escherichia coli*, *Listeria monocytogenes*, *Pseudomonas aeruginosa*, *Salmonella* spp., and *S. aureus* [55–58], or as inhibitors of total mesophilic, mould, and *Enterobacteriaceae* counts when applied to different foods [59]. However, there is a lack of studies investigating the influence of the addition of parsley, oregano, thyme, cinnamon, nettle, spinach, and carob on the microbiological quality of fresh pasta. From the present study, it is evident that cinammon and carob have the best potential as functional ingredients for fresh pasta production, especially in terms of microbiological quality and safety. However, the level of primary contamination of these and other desired ingredients for fresh pasta production must be taken into account.

4. Conclusions

In order to expand the range of fresh pasta with health benefits and a longer shelf life without freezing, the addition of various plant materials has been considered. The presence of plant ingredients in fresh pasta changed the cooking loss and textural characteristics of cooked pasta. Furthermore, the sensory attributes of pasta were affected due to the incorporation of materials that are pigment carriers and common spices with characteristic aromas. However, the listed changes did not impair the technological quality or acceptability of enriched pasta. By the 4th day of storage, the addition of plant materials had reduced the number of all tested microorganisms compared to the control sample. The positive effect on the reduction of microorganisms' count in pasta samples was maintained until the 8th day for all tested microorganisms except mold. This speaks in favour of plant ingredient effectiveness in the preservation of fresh pasta quality while stored in refrigerating conditions. The best quality in terms of technological, sensory, and microbiological aspects was achieved for cinnamon-enriched pasta. Although used plant materials are promising natural preservatives for pasta production, it is necessary to subject them to a decontamination process prior to usage. Further research should be conducted to investigate the antioxidant potential, starch digestibility, and glycaemic response of plant-enriched pasta.

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