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Fermented Cashew Nut Cheese Alternative Supplemented with *Chondrus crispus* and *Porphyra* sp.

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Abstract: This study is aimed at the development of a fermented cashew nut cheese alternative supplemented with *Chondrus crispus* and *Porphyra* sp. and the evaluation of the impact of seaweed supplementation through analysis of physicochemical, microbiological, and organoleptic properties of the developed food products. The total lipid content decreased with the supplementation with seaweeds. Crude protein content also slightly decreased, while elemental analysis showed that mineral and trace element (Ca, K, Mg, Na, Fe, I, Se, and Zn) content increased when *C. crispus* was added to the paste. The analyses of color and textural (TPA) attributes showed that these were significantly influenced by adding seaweeds to the cashew paste. Generally, the microbiological results comply with the different European guidelines for assessing the microbiological safety of ready-to-eat foods placed on the market, except for aerobic mesophilic bacteria and marine agar counts. Flash Profile analysis allowed for distinguishing sample attributes, showing an increased flavor complexity of the plant-based cheese alternatives supplemented with seaweeds. Overall, the study indicates that seaweed enrichment mainly influenced the physicochemical and sensory characteristics of plant-based cheese alternatives.

Keywords: plant-based cheese alternatives; seaweed supplementation; *Chondrus crispus*; *Porphyra* sp.; physicochemical characteristics; microbiota; sensory characteristics

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Citation: Campos, B.M.; Moreira-Leite, B.; Salgado, A.; Ramalho, E.; Marmelo, I.; Malfeito-Ferreira, M.; Sousa, P.; Diniz, M.S.; Mata, P. Fermented Cashew Nut Cheese Alternative Supplemented with *Chondrus crispus* and *Porphyra* sp. *Appl. Sci.* **2024**, *14*, 11082. https://doi.org/10.3390/app142311082

Academic Editors: Małgorzata Ziarno, Roberto Nigro, Marianna Gallo, Francesca Passannanti and Sara Sáez-Orviz

Received: 20 October 2024
Revised: 21 November 2024
Accepted: 26 November 2024
Published: 28 November 2024

1. Introduction

The demand for various plant-based cheese alternatives (PBCAs) is gradually gaining importance in the food market [1]. However, this market is in its beginning and needs to progress rapidly [2]. In 2020, the COVID-19 pandemic accelerated this process, making consumers re-evaluate their lifestyles and move towards plant-based diets [1]. This trend is justified by concerns related to the sustainable production of food, especially proteins [2], and/or to ethical reasons [3], such as animal welfare [4,5]. Health-related issues, such as food allergies and/or intolerances, also explain the increased consumption of plant-based dairy alternatives [6,7]. Some authors even suggest accelerated growth is driven by increased allergenicity toward cow's milk and lactose intolerance [1,5,8]. Furthermore, commonly reported motivations to adopt plant-based diets also included sensory/taste/disgust aspects and weight loss [6].

The main challenges associated with the development of plant-based alternatives to dairy products are to create products simultaneously innovative, sustainable, nutritious, and having organoleptic quality [3].

PBCAs are oil-in-water emulsions containing proteins, stabilizers, emulsifiers, flavors (cultures or nutritional yeast), food colorings, and preservatives. Usually, all the ingredients are blended to imitate the cheese's appearance and consistency [1]. According to Saraco and Blaxland [9], the primary ingredients used in dairy-free alternatives to cheeses found in the UK were coconut oil, almonds and/or cashew nuts, palm oil, rice, soya, and sunflower oil. Among these ingredients, the cashew nut (*Anacardium occidentale* L.) is one of the main plant-based sources of protein used in cheese alternatives [3].

In cashew nut-based cheese alternatives, about 50% of the weight is constituted by cashew nuts, followed by water and lemon juice, and sometimes added natural flavorings and phycocolloids such as agar-agar [10]. In other cases, PBCAs are coconut oil-based and are formulated using a low percentage of almonds and cashews along with a high coconut oil content [3]. Usually, cashew nut-based products present the highest protein levels and the lowest sodium and saturated fat contents [5]. For this reason, the replacement of dairy cheese with cashew nut-based options can help reduce the intake of salt and total fats, playing a special role in replacing the intake of saturated fats with unsaturated ones, thereby providing health benefits to consumers [10].

This study aimed to develop fermented cashew nut cheese alternatives (FCNCAs) supplemented with Irish Moss (*Chondrus crispus*) and Nori (*Porphyra* sp.) seaweeds. Other objectives included the characterization of the impact of seaweed supplementation in terms of their physicochemical, color, textural, microbiological, and sensory properties. Furthermore, according to the authors, no research has addressed the production of fermented cashew nut cheese alternatives with these characteristics. Thus, this is the first study conducting a comprehensive assessment of a plant-based cheese alternative supplemented with seaweeds.

2. Materials and Methods

2.1. Preparation of FCNCAs Supplemented with Seaweeds and Sampling Strategies

The experiments utilized dehydrated red seaweeds from the division Rhodophyta: Chondrus crispus (Stackhouse, 1797), commonly known as Irish Moss (order Gigartinales, family Gigartinaceae), and *Porphyra* sp. (C.A. Agardh, 1824), known as Nori (order Bangiales, family Bangiaceae). These seaweeds were harvested from Atlantic coastal waters and industrially air-dried (ALGAplus, Ltd., Ílhavo, Portugal). They were mechanically ground into small pieces using a grinder (Orbegozo BV 9600, Murcia, Spain), resulting in particle sizes of 2.87 ± 0.99 mm for *C. crispus* and 1.47 ± 0.49 mm for *Porphyra* sp. Then, they underwent UV irradiation (DNA/RNA UV-Cleaner Box, UVC/T-AR, Biosan, Riga, Latvia) for 48 h to prevent contamination, were vacuum-sealed (Sammic SU-316G, Azkoitia, Spain), and stored at room temperature for three days before being used to prepare FCNCAs, as detailed below.

Cashew nuts (Alesto®, Neckarsulm, Germany) were soaked in water for approximately 12 h at 6–7 °C. After soaking, they were boiled in water (~88 °C) for about 2 min, then drained and processed in a Thermomix® TM5 (Vorwerk, Wuppertal, Germany) at speed 8 during 3 min until a thick paste was formed. The paste was then mixed with 3% nutritional yeast flakes enriched with vitamin B12 (Marigold Health Foods® Engevita, London, UK), 3% unpasteurized white miso paste (Clearspring®, London, UK), and mineral water (Luso®, Vacariça, Portugal) and mixed until a thick paste was achieved. Then, 0.1% of Advanced Acidophilus Plus (Solgar®, Lisboa, Portugal), containing Lactobacillus acidophilus LA-5® (250 million organisms) and Bifidobacterium lactis BB-12® (250 million organisms), was added. The resulting mixture was transferred to a cheesecloth and pressed to remove excess water and/or fat.

Two percent of the selected seaweed (*Chondrus crispus* or *Porphyra* sp.) was incorporated into the paste. This mixture was then shaped using steel molds (Ø10 cm and 6–6.5 cm in height), placed in a hermetically sealed container, and stored at 6–7 °C. After two days, the product was removed from the molds, and the entire surface was sprinkled with 2% refined salt (Saldomar®, Olhão, Portugal). The fermented cashew nut cheese alternatives were then dried in an oven at 40 °C for 30 h (TS 9135, Termaks AS®, Bergen, Norway). Finally, the FCNCAs, uncovered, were ripened in a refrigerator (7.25 ± 1.50 °C at 59.17 ± 14.52% RH) (Thermo Meter, Max-Min Thermo Hygro TA318, KTJ®, Shenzhen, China), with daily flipping for 15 days.

Three different types of FCNCAs were manufactured: fermented cashew nut cheese alternative control (FCNCA-C), fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC), and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) (Figure 1).

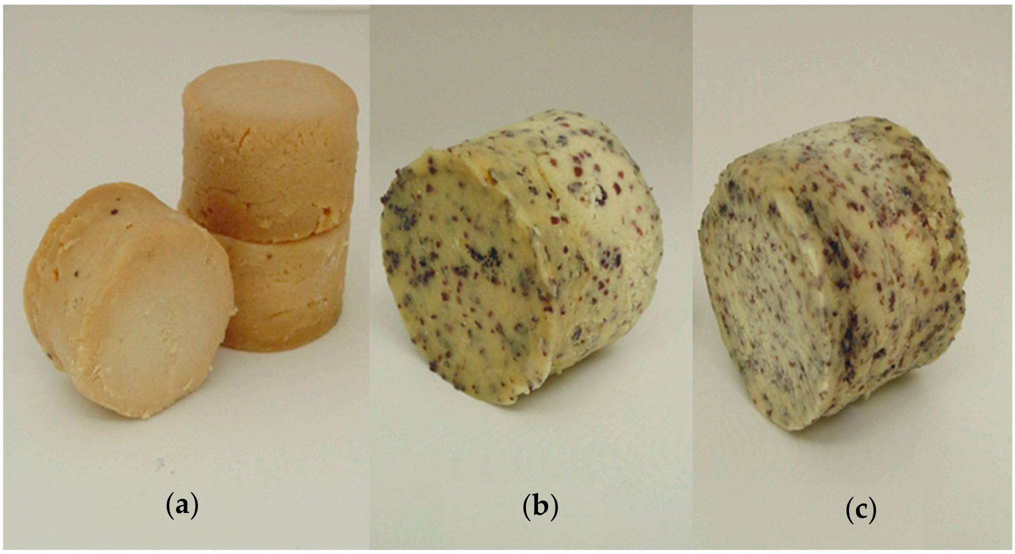

Figure 1. Representative images of the fermented cashew nut cheese alternatives (FCNCAs) after 15 days of ripening. From left to right: (**a**) fermented cashew nut cheese alternative control (FCNCA-C); (**b**) fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and (**c**) fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P).

After manufacturing, FCNCAs were stored according to the requirements of subsequent analyses. Some were vacuum-packed (Sammic SU-316, Azkoitia, Spain) in labeled polypropylene bags (90 µm, 180 mm × 300 mm, PA/PE, Sammic, Azkoitia, Spain); part of these were refrigerated (4–6 °C) and another part frozen (−18 °C) until further analysis. The remaining FCNCAs were freeze-dried in a laboratory freeze-dryer (ScanVac Cool Safe 4 L, LaboGene, Lillerød, Denmark), operating at a working temperature of −50 °C under a pressure of 0.0005–0.002 mBar, for 48 h. Subsequently, these were also vacuum-packed in labeled polypropylene bags and stored at −18 °C until further analysis.

The experimental design and general sampling strategy of the FCNCAs are summarized in Figure 2.

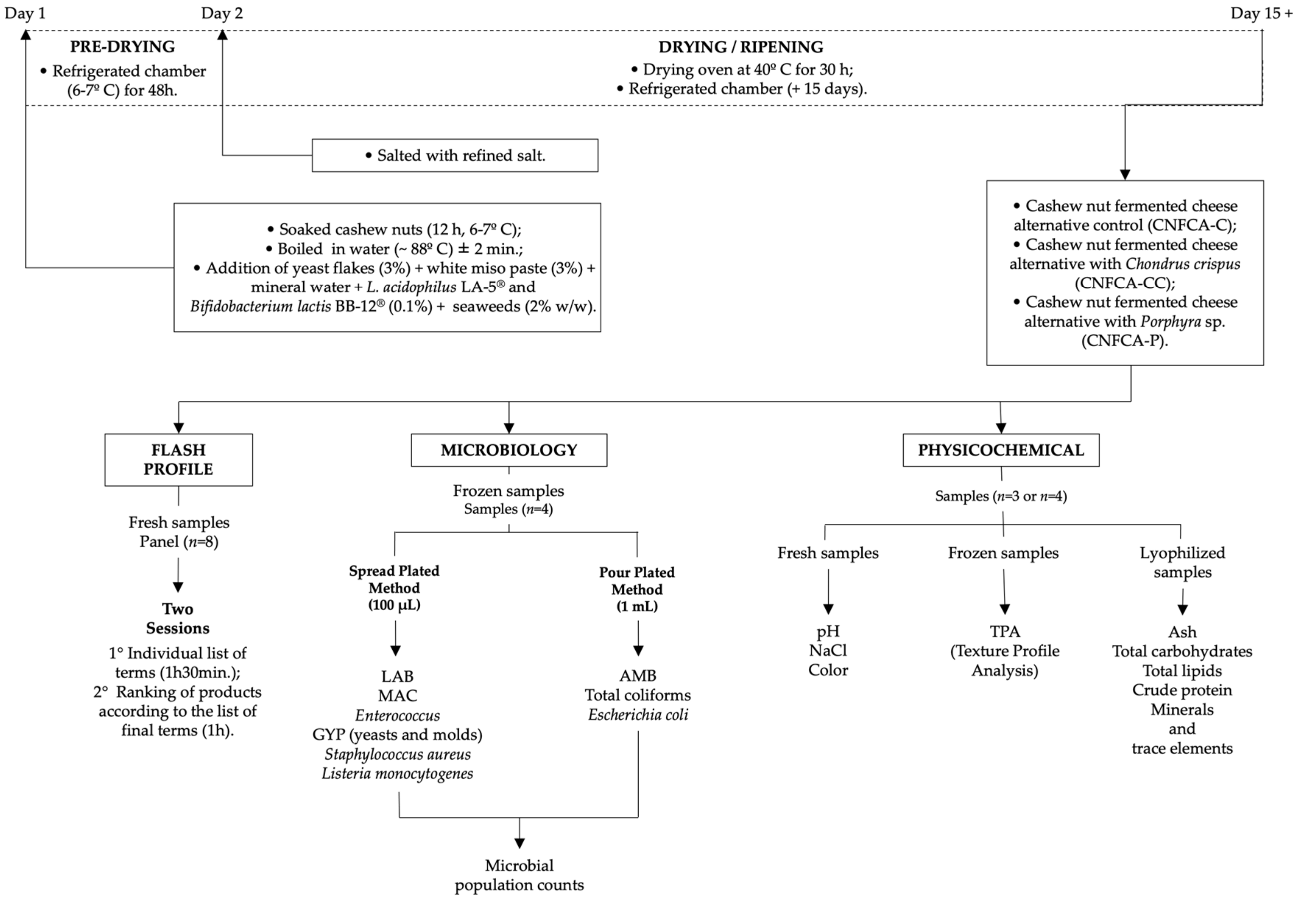

Figure 2. Flowchart of the production and analysis processes of the fermented cashew nut cheese alternatives (FCNCAs).

2.2. Physicochemical Analysis

2.2.1. Total Solid and Moisture Content

Total solids (TSs) were determined gravimetrically by measuring the weight loss of refrigerated samples (3 g) dried in an oven (TS 9135, Termaks AS®, Bergen, Norway) at 102 °C, using 20 g of sea sand (PanReac AppliChem ITW Reagents, Barcelona, Spain) until a constant weight was achieved. Then, TSs and moisture were calculated and expressed as percentage of wet weight (% WW).

2.2.2. Ash Content

Ash content (AC) was determined gravimetrically after the combustion of lyophilized samples (5 g) at 550 °C in a laboratory muffle furnace (LV 15/11/P320, Nabertherm GmbH, Bremen, Germany) until a constant weight was achieved. The ashes were cooled in a desiccator for at least one hour before weighing. The AC was calculated as the residue remaining after combustion and expressed as a percentage of dry weight (% DW).

2.2.3. NaCl and pH

Ten grams (10 g) of refrigerated samples were homogenized in 90 mL of Milli-Q water at 70 °C, and NaCl content was measured using a Digital Salinity-615 Salt Content Meter 0~199.9 ppt (Yieryi, Shenzhen, China). For pH determination, 20 g of refrigerated samples were blended with 12 mL of distilled water [11]. The pH was then measured using a pH meter (Nahita Model 903, Auxilab S.L., Beriáin, Spain) equipped with a glass electrode (XS Sensor Food S7, XS Instruments, Carpi, Italy).

2.2.4. Total Lipid Content

The gravimetric lipid assay was carried out according to the procedures previously described by Kumari et al. [12]. Briefly, 500 mg of lyophilized sample was mixed with 3.0 mL of a chloroform/methanol/50 mM phosphate buffer solution (Honeywell, Offenbach am Main, Germany; Fisher Scientific, Loughborough, UK) in a 2:1:0.8 (*v*/*v*/*v*) ratio. The mixture was vortexed for about 1 min and centrifuged (Domel, Centric 150, Otoki, Slovenia) at 2057× *g* for 15 min. Next, the residues were re-extracted three times with 2.0 mL of chloroform/methanol/buffer solution (1:1:0.8, *v*/*v*/*v*) and centrifuged as stated above. The supernatants were then combined, filtered, washed with 2.0 mL of 50 mM phosphate buffer, and centrifuged again at 2057× g for 5 min. Finally, the organic phase was dried under a regular nitrogen flow. To validate the methodology, the NIST Standard Reference Material® 3232—Kelp powder (*Thallus laminariae*) was used. The total lipid content was weighed (Radwag®, Model PS 450/X, Bracka, Poland) and expressed as a percentage of dry weight (% DW).

2.2.5. Crude Protein Content

Crude protein content was determined using an FP-528 combustion nitrogen (N) analyzer (LECO Corporation, St. Joseph, MI, USA). In this process, a pre-weighted freeze-dried sample (100 mg) was placed into the loading head of the combustion analyzer, sealed, and flushed with pure oxygen. The N covalently bound in the sample was converted to nitrogen gas (N_2) and measured using a thermal-conductivity cell. A calibration standard curve was built with ethylenediaminetetraacetic acid (EDTA, LECO 502-896, St. Joseph, MI, USA), and an air-blank was included for accuracy. Protein content was calculated by multiplying the measured nitrogen (N_2) content (%) by a conversion factor of 5.30 [13]. Results were expressed as a percentage of dry weight (% DW).

2.2.6. Elemental Analysis

In brief, 250 mg of samples were digested in 5 mL of nitric acid 65% (Merck, KGaA, Darmstadt, Germany) and 1.0 mL of hydrochloric acid 37% (Honeywell, Offenbach am Main, Germany) for 48 h using 15 mL Falcon tubes (Deltalab, Barcelona, Spain). The mixture was transferred to a fluoropolymer PFA (perfluoroalkoxy alkanes) microwave vessel, sealed, and heated on a digital dry bath (AccuBlock™, Labnet International, Inc., NJ, USA) at 100 °C for 24 h until complete digestion. After cooling, 100 µL of hydrogen peroxide 30% (Sigma-Aldrich/Merck, Darmstadt, HE, Germany) was added to each sample, vortexed, filtered, and diluted to a final volume of 10 mL. Blanks were prepared with ultrapure water under identical digestion conditions. Elemental analysis was performed by Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES) using an Ultima model (Horiba Jobin Yvon, Montpellier, France). Calibration curves were constructed for all elements. The minerals were expressed as $g \cdot kg^{-1}$ DW and trace elements as $mg \cdot kg^{-1}$ DW. The methodology was validated by using certified reference materials from the National Institute of Standards and Technology (NIST), namely, Kelp powder (*Thallus laminariae*) Standard Reference Material® (CRM) 3232.

2.2.7. Color

The FCNCAs were sliced into 1 cm thick pieces, and color parameters were measured in six distinct areas on the surface of each slice using a PCE-CSM 4 chroma meter (PCE Instruments™, Southampton, UK), with light source D65 (standard daylight) through coordinates CIELAB: L^* (degree of lightness perceived, $L^* = 0$, black, $L^* = 100$, diffuse white), a^* (degree of redness, $a^* > 0$, or greenness, $a^* < 0$), and b^* (degree of yellowness, $b^* > 0$, or blueness, $b^* < 0$). The colorimeter was calibrated using a standard white reference tile with the following values: $L^* = 93.97$; $a^* = -0.88$ and $b^* = 1.21$.

2.2.8. Texture Profile Analysis (TPA)

The texture profile analysis (TPA) was performed using a CT3 Texture Analyzer following a two-bite compression test with a TA4/1000-cylinder probe (Ø 38.1 mm, 20 mm in height) and a 4.5 kg force load cell (Brookfield, New York, NY, USA). The pre-test and post-test speed were set to 2.0 mm/s; trigger force, 2.0 g; compression, 20%; and time pause between cycles, 5 s. Defrosted FCNCAs were cut into cubes (20 × 20 × 20 mm) and allowed to equilibrate at room temperature (~22 °C) for about two hours. From the compression, curves were obtained, and parameters such as hardness (N), adhesiveness (J), cohesiveness (—), springiness (mm), and gumminess (N) were derived using the software TexturePro CT Software 1.6 (AMETEK Brookfield, New York, NY, USA).

2.3. Microbial Load

Frozen samples were prepared in a horizontal laminar airflow cabinet (Aeolus H, Telstar®, Terrasa, Spain) for microbial analysis. Depending on the test, either 10 g or 25 g of each sample was weighed (Radwag®, Model PS 450/X, Bracka, Poland), taken aseptically, placed in a BagLight Poly-Silk sterile blender bag (Interscience, Saint-Nom-la-Bretèche, France), and homogenized in 90 mL of Ringer's solution (Biokar Diagnostics, Pantin, France) during 90 s in a stomacher apparatus (BagMixer® 400 P, Interscience, Saint-Nom-la-Bretèche, France). Serial dilutions were prepared in sterile ¼ strength Ringer solution and either spread-plated (100 µL) or dispersed using the pour plate method (1 mL) in Ø90 mm Petri dishes (Frilabo, Maia, Portugal) for viable counts.

The following microbiological analyses were conducted: (a) total aerobic mesophilic bacteria (AMB) on Plate Count Agar (PCA) (Biokar Diagnostics, Pantin, France), incubated at 30 °C for 72 h; (b) Lactic Acid Bacteria (LAB) on De Man, Rogosa, and Sharpe agar (MRS broth) (Biokar Diagnostics, Pantin, France), acidified to a final pH of 5.4 ± 0.1 with acetic acid (Sigma-Aldrich/Merck, Darmstadt, HE, Germany) and incubated at 30 °C for 72 h; (c) *Enterococcus* on Compass® *Enterococcus* agar (Biokar Diagnostics, Pantin, France), incubated at 44 °C for 24 h; (d) total coliforms and *Escherichia coli* on Compass® ECC agar (Biokar Diagnostics, Pantin, France), incubated, respectively, at 37 °C and 44 °C for 24 h; (e) coagulase-positive staphylococci (*Staphylococcus aureus*) on Baird-Parker RPF agar (BP) (Biokar Diagnostics, Pantin, France), incubated at 37 °C for 48 h; (f) yeasts and molds on Chloramphenicol Glucose Agar (CGA) (Biokar Diagnostics, Pantin, France), incubated at 25 °C for 5 days; (g) marine bacteria on marine agar (Condalab, Madrid, Spain), incubated at 20–25 °C for 72 h; (h) *Salmonella* spp. on Buffered Peptone Water (BPW), Rappaport-Vassiliadis Soja (RVS) broth (RAP), Muller-Kauffmann Tetrathionate-Novobiocin (MKTTN) broth, Xylose Ly sine Desoxycholate (XLD) agar, and Brilliant Green Agar (BGA) (Biokar Diagnostics, Pantin, France), incubated at 37 °C for 5 days; (i) *Listeria monocytogenes* on Half-Fraser broth, Fraser broth, and Palcam agar (Biokar Diagnostics, Pantin, France), incubated at 37 °C for 5 days.

After counting, the means and standard deviations (SDs) were calculated and expressed in log CFU$\cdot$g^{-1}. The results were interpreted according to the United Kingdom's Health Protection Agency guidelines for assessing the microbiological safety of ready-to-eat foods placed on the market [14], the INSA Portuguese guidelines [15], and the European Commission Regulation (EC) No. 2073/2005 [16]. The test methods used for the various microbiological parameters are provided in the Supplementary Materials (Table S1).

2.4. Flash Profile (FP)

The panel comprised eight trained panel members (seven women and one man), with 50% aged between 18 and 30 and 50% aged between 30 and 65 years old. Panel members were recruited from among the students and academic staff of the Chemistry Department, NOVA School of Science and Technology, NOVA University, Lisbon, Portugal, in 2020.

The FP test consisted of two sessions, where the samples were coded with three-digit random numbers and presented simultaneously to the panel members. The members were instructed to rinse their mouths with tap water at room temperature before and between each sample.

During the first session (1 h 30 min), the panel members were given a brief outline of the procedures and asked to individually list the attributes they felt that best describe the differences between each sample (appearance, aroma, flavor, texture, and after-taste). They were instructed to avoid the use of hedonic terms. Each panelist was given the opportunity to compare his own list of sensory attributes with a joint list of attributes selected by all members of the panel. They were then allowed to modify their list by adding, renaming, or deleting any of their own attributes.

In the second session, panelists were asked to rank the FCNCAs on their own at-tributes according to differences in intensity. For each sensory attribute, a line scale was used, anchored at both extremes with the lowest and highest degrees of intensity, and ties were permitted. The session lasted about 1 h. In both cases, all sessions were conducted individually.

2.5. Statistical Analysis

A non-parametric Mann–Whitney U test was carried out to assess any significant differences between the means. In all cases, statistical significance was considered at $p < 0.05$. All statistical analyses were carried out using STATISTICA software (version 8.0, StatSoft Inc., Tulsa, OK, USA). All data were expressed as mean ± standard deviation (SD) and are reported on a dry matter basis unless stated otherwise.

Flash Profile (FP) results were analyzed through Generalized Procrustes Analysis (GPA) using Microsoft Office Excel add-in software, XLSTAT version 2022 (Addinsoft, New York, NY, USA) to obtain the optimal configuration in individual scaling data [17], thus minimizing the differences between the panelists. After the GPA consensus configuration, the FP data generate biplot maps showing the products' differences and similarities of the products according to the graphic interpretation.

3. Results and Discussion

3.1. Physicochemical Characterization of FCNCAs

Table 1 shows the FCNCAs physicochemical parameters (mean ± SD) and Table S2 (Supplementary Materials) the p-values.

Table 1. Physicochemical characterization of fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) *.

Physicochemical Parameters	**Control**	**Supplemented Products**	
	FCNCA-C	**FCNCA-CC**	**FCNCA-P**
Total solids (% WW)	64.63 ± 0.88 ^{a}	67.96 ± 0.53 ^{b}	69.79 ± 0.59 ^{c}
Moisture (% WW)	35.37 ± 0.88 ^{a}	32.04 ± 0.53 ^{b}	30.21 ± 0.59 ^{c}
Ash (% DW)	2.81 ± 0.08 ^{a}	3.63 ± 0.15 ^{b}	3.12 ± 0.09 ^{c}
NaCl (g/100 g)	2.67 ± 0.21 ^{a}	2.50 ± 0.10 ^{a}	2.43 ± 0.15 ^{a}
pH	5.27 ± 0.12 ^{a}	5.07 ± 0.15 ^{a,b}	5.07 ± 0.06 ^{b}
Lipids (% DW)	37.45 ± 0.44 ^{a}	34.51 ± 3.85 ^{b}	32.91 ± 0.16 ^{b}
Crude protein (% DW)	18.92 ± 0.00 ^{a}	18.26 ± 0.45 ^{b}	18.73 ± 0.20 ^{c}

* Mean ± SD (n = 3) except for crude protein (n = 4). Different letters indicate significant differences among treatments ($p < 0.05$).

3.2. Total Solid and Moisture Content

Total solid (TS) values ranged from 65 to 70% WW, and statistical analyses revealed significant differences among all samples ($p < 0.05$). The TS content of supplemented products increased by 3–5% WW. In general, the values found are higher than those reported for formulated processed dairy cheese (44.25%) or formulations with different ratios (2.5–10%) of a vegetable blend, where values vary from 47 to 53% [18].

The moisture content ranged from 30 to 35% WW, with significant differences between all samples ($p < 0.05$). These values are lower than those reported by Chen et al. [19] for fermented cashew cheese brie (43.4%) and by Grasso et al. [20] for commercial plant-based block-style alternative products (46–54%).

3.3. Ash Content

The ash content (AC) shows significant differences among all samples ($p = 0.0495$). When seaweeds are added, the AC of the samples increases from 2.81% (FCNCA-C) to 3.12% (FCNCA-P) and to 3.63% (FCNCA-CC). In fact, as reported by Campos et al. [20], *C. crispus* has a higher AC content (26.0%) than *Porphyra* sp. (13.9%). Also, the AC of dried cashew nuts reported by other researchers ranges from 2.02% [21] to 2.7% [22], within the range found in the present study.

3.4. NaCl and pH

NaCl content ranged from 2.43 to 2.67 g/100 g; no significant differences were found for FCNCAs ($p > 0.05$). Other studies reported lower values for plant-based alternatives, namely, 0.6 (0.5–0.6) g/100 g (expressed as median (min.–max.)) in cashew nut-based cheese alternative [10] or 1.25 ± 0.115 (g/100 g) for plant-based alternatives made of almond, sunflower, and cashew [7]. The determined values are within the levels of NaCl for dairy cheeses produced by rennet coagulation, namely, 0.7–4 g/100 g [23].

The pH ranged from 5.07 to 5.27, showing a significant difference between FCNCA-C and FCNCA-P ($p = 0.0431$). Despite this, in all FCNCAs, the accumulation of organic acids was not enough to achieve a final pH ≤ 4.4, which is a safe threshold to minimize the growth of *L. monocytogenes* [24,25].

3.5. Total Lipid Content

Total lipid (TL) content differs significantly for FCNCA-C vs. FCNCA-CC and FCNCA-C vs. FCNCA-P ($p = 0.0495$). The cashew nut kernels are rich in lipids [26], showing a high fat content in the whole seed (43–50%) [27]. Other researchers reported ca. 66 g/100 g of DW in raw cashew nut kernels [28]. In this study, the TL content of FCNCAs ranged from 33 to 37%, decreasing 3–5% when seaweeds were added to the matrix—this is likely due to the very low lipid content of seaweeds [20,29].

3.6. Crude Protein Content

Crude protein contents ranged from 18.26 to 18.92% and showed significant differences between all samples ($p = 0.0180$), although the values are similar, with less than 1% of difference between them. Although red seaweeds have high levels of protein (14–47 g/100 g of dry weight) [30,31], their supplementation does not contribute to protein improvement in FCNCAs, as cashew nuts have a similar protein content (21.3%) [32].

Proteins have many functionalities in foods, namely, solubility, gelling, foaming, and flavor creation. Plant proteins have a different structure, composition, and food functionality than animal proteins [33]. In general, alternative cashew nut-based cheese products were considered energy-dense and presented a protein median of 11 g/100 g, representing a good source of proteins [10].

Nowadays, developed countries raise health and environmental concerns about the consumption of animal protein. A high animal protein intake has been associated with an increased risk of several diseases (e.g., cardiovascular, carcinogenic, diabetes, and obesity) [34]. Therefore, nut kernels can be an important source of protein for plant-based diets [35]. The replacement of dairy cheese with cashew nut-based options was suggested to moderate the high protein intake by Spaniard consumers, for example, since they have an excessive protein intake [10].

3.7. Elemental Analysis

The concentrations of minerals and trace elements for seaweed and the FCNCAs are presented in Table 2 and the *p*-values of supplemented products in the Supplementary Materials (Table S3).

Table 2. Contents for minerals and trace elements in each seaweed analyzed (Irish Moss *(Chondrus crispus*) and Nori (*Porphyra* sp.)) and fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) *.

Minerals and Trace Elements	***C. crispus***	***Porphyra* sp.**	**FCNCA-C**	**FCNCA-CC**	**FCNCA-P**	**Certified Values *****
Ca ($g{\cdot}kg^{-1}$ DW)	4.29 ± 0.17^{A}	1.77 ± 0.05^{B}	0.81 ± 0.09^{a}	1.07 ± 0.06^{b}	1.77 ± 0.19^{ab}	12.35 ± 0.22
K ($g{\cdot}kg^{-1}$ DW)	36.24 ± 1.09^{A}	16.55 ± 0.48^{B}	2.59 ± 0.24^{a}	3.79 ± 0.31^{b}	2.79 ± 1.05^{ab}	75.18 ± 1.66
Mg ($g{\cdot}kg^{-1}$ DW)	6.82 ± 0.07^{A}	4.26 ± 0.12^{B}	3.13 ± 0.19^{a}	3.51 ± 0.15^{b}	3.19 ± 0.50^{ab}	6.08 ± 0.21
Na ($g{\cdot}kg^{-1}$ DW)	31.81 ± 0.58^{A}	22.40 ± 0.95^{B}	6.19 ± 0.24^{a}	9.53 ± 0.18^{b}	7.38 ± 1.10^{c}	16.56 ± 0.49
P ($g{\cdot}kg^{-1}$ DW)	1.70 ± 0.05^{A}	2.31 ± 0.09^{B}	7.59 ± 0.41^{a}	6.64 ± 0.65^{b}	7.98 ± 0.66^{ab}	4.58 ± 0.48
Fe ($mg{\cdot}kg^{-1}$ DW)	123.95 ± 5.8^{A}	79.96 ± 1.73^{B}	49.48 ± 2.14^{a}	60.14 ± 0.21^{b}	48.21 ± 2.97^{a}	661.03 ± 33.11
I ($mg{\cdot}kg^{-1}$ DW)	29.33 ± 0.11^{A}	15.80 ± 0.82^{B}	<LOQ **	1.26 ± 0.30	<LOQ **	918.05 ± 49.52
Mn ($mg{\cdot}kg^{-1}$ DW)	26.18 ± 0.88^{A}	20.77 ± 0.79^{B}	15.03 ± 0.18^{a}	15.84 ± 1.29^{a}	14.88 ± 1.31^{a}	23.90 ± 1.81
Se ($mg{\cdot}kg^{-1}$ DW)	0.84 ± 0.10^{A}	2.49 ± 0.10^{B}	2.05 ± 0.02^{a}	2.54 ± 0.33^{b}	2.15 ± 0.19^{ab}	n.a.
Zn ($mg{\cdot}kg^{-1}$ DW)	66.24 ± 3.97^{A}	48.05 ± 1.89^{B}	40.13 ± 2.52^{a}	65.76 ± 2.51^{b}	46.78 ± 6.55^{a}	26.52 ± 0.63

* Mean ± standard deviation ($n = 3$). Different uppercase letters indicate significant differences in the mineral composition between seaweed species, while lowercase letters indicate significant differences among treatments ($p < 0.05$). ** Below the Limit of Quantification. *** Certified reference materials (CRMs) from the National Institute of Standards and Technology (NIST): Kelp powder (*Thallus laminariae*) Standard Reference Material® (CRM) 3232. n.a. (not analyzed as no values are available in the Standard Reference Material® (CRM) 3232 analysis certificate).

For the FCNCAs, calcium (Ca) ranged from 0.81 to 1.77 $g{\cdot}kg^{-1}$, showing a significant difference between FCNCA-C and FCNCA-CC ($p = 0.0495$). The results also show that *C. crispus* has higher levels of Ca (4.29 $g{\cdot}kg^{-1}$) than *Porphyra* sp. (1.77 $g{\cdot}kg^{-1}$,), and this has a decisive impact on the paste of FCNCA-CC (1.07 $g{\cdot}kg^{-1}$). As Boukid et al. [1] reported, plant-based products have lower calcium content when compared to dairy products. However, the values determined in this study are higher than those observed for cashew cheese brie (0.41 $g{\cdot}kg^{-1}$) without any type of supplementation [19].

Potassium (K) content in FCNCAs ranged from 2.59 to 3.79 $g{\cdot}kg^{-1}$, showing significant differences between FCNCA-C and FCNCA-CC ($p = 0.0495$) and reflecting the higher K content found in *C. crispus*. When compared to fermented cashew cheese brie (1.76 $g{\cdot}kg^{-1}$) [19], all FCNCA levels are higher, especially when supplemented with *C. crispus* (3.79 $g{\cdot}kg^{-1}$).

Magnesium (Mg) ranges from 3.13 to 3.51 $g{\cdot}kg^{-1}$, showing a significant difference between FCNCA-C and FCNCA-CC ($p = 0.0495$), with *C. crispus* exhibiting a positive effect when added to the paste, which is explained by the obtained results for the seaweeds, namely, 4.26 for *Porphyra* sp. and 6.82 for *C. crispus*. Magnesium (Mg) deficiency is common in humans [36], playing an important role in the pathogenesis of ischemic heart disease, congestive heart failure, cardiac arrhythmias, and hypertension, among others [37], and as such is a critical mineral in the human body [38].

Sodium (Na) ranged from 6.19 to 9.53 $g{\cdot}kg^{-1}$ and showed significant differences between all samples ($p = 0.0495$), with supplemented products presenting higher values than controls, which are directly related to the contents determined for seaweeds, namely, 31.81 $g{\cdot}kg^{-1}$ for *C. crispus* and 22.40 $g{\cdot}kg^{-1}$ for *Porphyra* sp. The values obtained for controls (6.19 $g{\cdot}kg^{-1}$) are in agreement with values reported by Chen et al. [19] for cashew cheese brie (6.26 $g{\cdot}kg^{-1}$).

Phosphorus (P) ranged from 6.64 to 7.98 $g{\cdot}kg^{-1}$ and showed significant differences between FCNCA-C and FCNCA-CC ($p = 0.0495$), since the P levels measured in *C. crispus* are very low (1.70 $g{\cdot}kg^{-1}$).

Regarding trace elements, iron (Fe) levels range from 48.21 to 60.14 mg·kg^{-1}, showing significant differences between FCNCA-C and FCNCA-CC and between FCNCA-CC and FCNCA-P (p = 0.0495). The high Fe content found in *C. crispus* (123.95 mg·kg^{-1}) significantly impacted the supplemented cheese, contrary to *Porphyra* sp. (79.96 mg·kg^{-1}). However, all values obtained for FCNCAs are higher than the value reported by Chen et al. [19] for cashew cheese brie (17 mg·kg^{-1}).

It was only possible to quantify the iodine (I) content for FCNCA-CC, which can be attributed to the higher iodine content of *C. crispus* (29.33 mg·kg^{-1}). The results for FCNCA-C and FCNCA-P are below the detection limit, and the low percentage (2%) of *Porphyra* sp. seaweed used for supplementation can justify this.

In a recent study carried out by Clegg et al. [7], of 109 cheese alternatives available in the UK, none of them were fortified with iodine, an essential nutrient required to produce thyroid hormones, which play a crucial role in growth mechanisms and the development of tissues [7,39].

Manganese (Mn) ranged from 14.88 to 15.84 mg·kg^{-1} and did not show significant differences among analyzed samples (p > 0.05).

Selenium (Se) ranges from 2.05 to 2.54 mg·kg^{-1}, showing a significant difference between FCNCA-C and FCNCA-CC (p = 0.0495). Although *Porphyra* sp. content is higher (2.49 mg·kg^{-1}) than that of *C. crispus* (0.84 mg·kg^{-1}), its addition to the paste had no enrichment effect, contrary to *C. crispus*. Selenium (Se) is an important oligoelement playing a crucial role in the antioxidant defense system due to its requirement by the Se-dependent GSH-Px, which is involved in cellular antioxidant protection [40].

Zinc (Zn) ranged from 40.13 to 65.76 mg·kg^{-1}, showing significant differences between FCNCA-C and FCNCA-CC and between FCNCA-CC and FCNCA-P (p = 0.0495), with FCNCA-CC presenting the best results, which are in agreement with the content found in *C. crispus* (66.24 mg·kg^{-1}).

3.8. Color

The color of FCNCAs was significantly influenced by adding seaweeds (Table 3). The p-values are presented in the Supplementary Materials (Table S4). Lightness (L*) is significantly different for FCNCA-C vs. FCNCA-CC, FCNCA-C vs. FCNCA-P (p = 0.0002), and FCNCA-CC vs. FCNCA-P (p = 0.0012). Fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) recorded the lowest value (38.30) for L*, followed by FCNCA-CC (43.77), with the highest value for FCNCA-C (53.33), i.e., the pigments from red seaweeds affected the lightness of FCNCAs, especially *Porphyra* sp.

Table 3. Color parameters of fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) *.

Parameters	Control	Supplemented Products	
	FCNCA-C	FCNCA-CC	FCNCA-P
L*	53.33 ± 1.91 ^{a}	43.77 ± 2.25 ^{b}	38.30 ± 2.21 ^{c}
a*	10.56 ± 0.47 ^{a}	3.19 ± 0.50 ^{b}	6.68 ± 0.94 ^{c}
b*	25.74 ± 0.46 ^{a}	15.62 ± 2.28 ^{b}	13.71 ± 1.43 ^{c}
C*	27.83 ± 0.52 ^{a}	15.95 ± 2.29 ^{b}	15.26 ± 1.50 ^{b}
WI	45.67 ± 1.88 ^{a}	41.55 ± 2.16 ^{b}	36.44 ± 2.46 ^{c}

* Values of color parameters (L* (lightness); a* (redness to greenness); b* (yellowness to blueness); and C* (chroma)) are presented as mean ± standard deviation (n = 4). Different letters indicate significant differences among treatments (p < 0.05).

The redness to greenness (a*) shows significant differences among all the samples (p = 0.0002). Fermented cashew nut cheese alternative with *C. crispus* (FCNCA-CC) exhibits the lowest value (3.19), followed by FCNCA-P (6.68), with the highest value for FCNCA-C (10.56). Seaweeds of the division Rhodophyta are a valuable source of chlorophyll *a* and *d*, phycobilins (allophycocyanin (APC), R-phycoerythrin (R-PE), R-phycocyanin (R-PC)), carotenoids (*α*-, and *β*-carotene), and xanthophylls (lutein) which influenced the color of FCNCAs [41,42].

C. crispus has a lower value of total pigment content (0.52 mg/g), with *β*-carotene (73.76%) as the main pigment, followed by chlorophyll *a* (26.17%) and lutein (0.061%) [43]. However, in dried form (DF) and after hydration treatment (HT), *β*-carotene is not detectable in *C. crispus*. At the same time, phycoerythrin (Δλ = 10) decreases from 528.3 ± 69.0 mg/Kg dry matter (DF) to 525.6 ± 74.2 mg/Kg dry matter (HT); phycocyanin (Δλ = 40) increases from 149.2 ± 22.8 mg/Kg dry matter (DF) to 232.5 ± 27.0 mg/Kg dry matter (HT); and lutein increases from not detected (DF) to 1.80 ± 0.17 mg/Kg dry matter (HT) [44]. In the case of *Porphyra* spp., in addition to phycobiliproteins such as phycoerythrin (PE) and phycocyanin (PC) pigments, chlorophylls *a* and *d* are also present, as well as several carotenoids (*β*-carotene, *α*-carotene, lutein, zeaxanthin, violaxanthin, and fucoxanthin) [45]. In the present case, the pigments of both seaweeds affect the redness to greenness (a^*) parameter, being the values for the FCNCAs closer to the greenish direction. Due to the high fat content and the lower polarity of chlorophylls (compared to phycobiliproteins), it is possible that there was better solubilization of chlorophylls in the cashew paste.

Yellowness to blueness (b^*) is positive in all FCNCAs and therefore in the yellow range of color values, but significant differences were found between FCNCA-C and FCNCA-CC, between FCNCA-C and FCNCA-P ($p = 0.0002$), and between FCNCA-CC and FCNCA-P ($p = 0.0494$). Fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) presents the lowest value (13.71), followed by FCNCA-CC (15.62), with the highest value by FCNCA-C (25.74). These values also result from the presence of the various seaweed pigments referred to, contributing to less yellowness.

Chroma (C^*) values, measuring color purity and intensity, reveal significant differences between FCNCA-C and FCNCA-CC and between FCNCA-C and FCNCA-P ($p = 0.0002$), with the lowest value being found in FCNCA-P (15.26), followed by FCNCA-CC (15.95), with the highest values determined in FCNCA-C (27.83), reflecting the effect of the various pigments present in the seaweeds.

Overall, FCNCA-C presents the highest value in all parameters. In general, FCNCAs exhibited mean levels of L^* (≤ 53), with component b^* predominant over component a^*, suggesting that the degree of lightness and yellowness mostly contributed to the color features of the FCNCAs, as well reported by other authors [46,47].

The whiteness index (WI) indicates the degree of whiteness and mathematically combines lightness (L^*) and yellowness to blueness (b^*) into a single term [48].

Results show that whiteness indices are proportional to lightness indices (FCNCA-C>FCNCA-CC>FCNCA-P). Therefore, fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) presents the lowest value (36.44), followed by FCNCA-CC (41.55), with the highest value by FCNCA-C (45.67), showing that a decrease in whiteness arose from the supplementation of seaweeds particles.

3.9. Texture Profile Analysis (TPA)

The textural properties of the FCNCAs are reported in Table 4 and *p*-values in the Supplementary Materials (Table S5). TPA results are difficult to discuss, as there is little or no data from other studies.

The hardness of the FCNCAs ranged from 6.01 to 9.69 N, showing statistical differences between FCNCA-C and FCNCA-CC ($p = 0.0065$), between FCNCA-C and FCNCA-P, and between FCNCA-CC and FCNCA-P ($p = 0.0039$). Hardness can be defined as the force required to attain a given deformation [49]. Increasing the moisture content has the opposite effect on hardness [50], and in fact, there is an inverse relationship between the hardness and moisture content of the FCNCAs.

An inverse relationship with lipid content can also be observed which can explain hardness differences. The lowest total lipids resulted in FCNCAs with higher hardness. A possible explanation for this might be that the fat in these alternative cheeses is mainly unsaturated. In fact, a study conducted by Devi and Khatkar [51] concluded that saturated fatty acids contribute to dough hardness, and fats rich in unsaturated fatty acids such as sunflower oil produced the softest cookie dough.

Table 4. TPA parameters (hardness (N), adhesiveness (J), cohesiveness (—), springiness (mm), and gumminess (N)) for fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) *.

Supplemented Products	Hardness (N)	Adhesiveness (J)	Cohesiveness (—)	Springiness (mm)	Gumminess (N)
FCNCA-C	6.01 ± 0.56^{a}	0.07 ± 0.06^{a}	0.33 ± 0.04^{a}	1.70 ± 0.25^{a}	2.00 ± 0.23^{a}
FCNCA-CC	7.90 ± 0.68^{b}	0.17 ± 0.09^{a}	0.37 ± 0.03^{a}	1.91 ± 0.30^{a}	2.93 ± 0.18^{b}
FCNCA-P	9.69 ± 0.66^{c}	0.03 ± 0.04^{a}	0.42 ± 0.02^{b}	2.29 ± 0.19^{b}	4.08 ± 0.45^{c}

* Mean ± standard deviation ($n = 4$). Different letters indicate significant differences among treatments ($p < 0.05$).

Adhesiveness ranged from 0.03 to 0.17 J but did not exhibit significant differences between samples ($p > 0.05$). The slightly higher value attributed to FCNCA-CC (0.17 J) can be due to the carrageen's polysaccharides in *C. crispus* [52,53].

Adhesiveness can be defined as the work necessary to overcome the attractive forces between the surface of the food and the surface of other materials with which the food comes into contact [49]. The positive low adhesiveness values show that FCNCAs are not very sticky or adhesive.

Cohesiveness values range from 0.33 to 0.42, showing significant differences between FCNCA-C and FCNCA-P ($p = 0.0035$) and between FCNCA-CC and FCNCA-P ($p = 0.0240$). Cohesiveness indicates the strength of the internal bonds making up the body of the product [54]. The mean cohesiveness values for FCNCA-P indicate that the structure is not easily disintegrated [55]. The fiber content can also justify the highest cohesiveness values for supplemented products. The reported insoluble dietary fiber content of Spanish seaweeds for *C. crispus* (12.04%) and for *Porphyra* sp. (19.22%) [56] is directly related to the values found in the present study. Other factors contributing to these differences may be the moisture and lipid contents.

Springiness (elasticity) is defined as the distance recovered by the sample during the time that elapsed between the end of the first compression and the start of the second one [49,57]. Springiness results revealed that the minimum (1.70 mm) was found in FCNCA-C, while the maximum value (2.29 mm) is for FCNCA-P. Significant differences between FCNCA-C and FCNCA-P ($p = 0.0103$) and between FCNCA-CC and FCNCA-P ($p = 0.0245$) were found.

Gumminess values ranged from 2.00 to 4.08 N, showing significant differences between all samples ($p = 0.0039$). Gumminess is the denseness that persists through chewing, or the energy needed to disintegrate a semisolid food until it is ready for swallowing [57]. Since supplemented products present higher values, more chews are required before swallowing. There is also a clear correlation between gumminess and hardness, cohesiveness, and springiness, as well as an inverse correlation with total lipid content and moisture.

3.10. Microbial Load

The microbiological evaluation of FCNCAs is presented in Table 5, and the *p*-values are in the Supplementary Materials (Table S6). The mean counts of *Enterococcus* exceed 10^4 or 4.0 log CFU·g^{-1} for all samples and do not show significant differences between them ($p > 0.005$).

Lactic Acid Bacteria (LAB) showed a significant difference for FCNCA-C vs. FCNCA-CC and FCNCA-CC vs. FCNCA-P ($p = 0.0139$). The LAB predominated over other microbiological groups, achieving the same counts for FCNCA-CC and FCNCA-P (5.48 log CFU·g^{-1}) and slightly exceeding by 1.08 log CFU·g^{-1} for FCNCA-C. In all cases, the values obtained were higher than the limits referred by Saraiva et al. [15]. These elevated counts can be related to the addition of Advanced Acidophilus Plus (Solgar®), which contained a large number of *Lactobacillus acidophilus* and *Bifidobacterium lactis*. In fact, Lactobacillus is a genus with important applications in food fermentation, as it is also capable of producing lactic acid due to the metabolism of sugars [58]. Apart from that, LAB is responsible for producing substances that improve flavor, texture, nutritional value, shelf-life, and safety of foods [59].

Table 5. Levels of *Enterococcus*, Lactic Acid Bacteria (LAB), aerobic mesophilic bacteria (AMB), marine agar counts (MACs), Glucose–Yeast–Peptone (GYP) molds and yeasts, *Escherichia coli*, total coliforms (TCs), *Staphylococcus aureus*, *Salmonella* spp., and *Listeria monocytogenes* for fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) *.

Microbiological Parameters (Log CFU·g^{-1})	**Control**	**Supplemented Products**	
	FCNCA-C	**FCNCA-CC**	**FCNCA-P**
Enterococcus	4.48 ± 000 ^{a}	4.48 ± 0.00 ^{a}	4.48 ± 0.00 ^{a}
LAB	6.56 ± 0.00 ^{a}	5.48 ± 0.00 ^{b}	5.48 ± 0.00 ^{b}
AMB	7.83 ± 0.86 ^{a}	7.36 ± 0.80 ^{b}	7.48 ± 0.00 ^{c}
MAC	<2 ^{a}	6.18 ± 1.63 ^{b}	6.08 ± 0.18 ^{b}
GYP (Molds)	<2 ^{a}	<2 ^{a}	<2 ^{a}
GYP (Yeasts)	3.76 ± 1.64 ^{a}	<2 ^{b}	<2 ^{b}
E. coli	2.11 ± 0.33 ^{a}	<1 ^{b}	1.9 ± 1.89 ^{a}
TC	0.7 ± 0.15 ^{a}	0.7 ± 0.15 ^{a}	2.57 ± 0.00 ^{b}
S. aureus	2.18 ± 1.63 ^{a}	2.54 ± 0.37 ^{a,b}	2.72 ± 0.42 ^{b}
Salmonella spp.	Absent in 25 g	Absent in 25 g	Absent in 25 g
L. monocytogenes	Absent in 25 g	Absent in 25 g	Absent in 25 g

* Counts are expressed as log CFU·g^{-1}. Mean ± standard deviation (n = 4). Different letters indicate significant differences among treatments. ($p < 0.05$).

Aerobic mesophilic bacteria (AMB) are the most represented group, ranging from 7.36 to 7.83 log CFU·g^{-1}. The statistical analyses revealed significant differences among all FCNCAs ($p < 0.05$). Usually, cashew nut or cashew nut products present a higher AMB activity. For example, Muniz et al. [60] reported 4.0 to 7.0 log CFU·g^{-1} for cashew nut, whereas Göçer and Koptagel [61] reported 8.84 ± 0.26 log CFU·g^{-1} for a cashew nut-based beverage fermented with kefir. According to Saraiva et al. [15], all FCNCAs are rated as unsatisfactory (>10^{7} or 7 log CFU·g^{-1}) (see Supplementary Materials, Table S7).

Counts on marine agar achieved similar values among FCNCA-CC (6.18 log CFU·g^{-1}) and FCNCA-P (6.08 log CFU·g^{-1}). Fermented cashew nut cheese alternative control (FCNCA-C) does not show microbiological activity (<2 log CFU·g^{-1}). A plausible justification for this is that the values for the supplemented products are due to supplementation with seaweeds.

Molds were deemed satisfactory, presenting the same values for all samples (<2 log CFU·g^{-1}), and did not show significant differences between them ($p > 0.05$). Some authors suggest that contamination of cashew nuts by molds can occur early in the field or during a prolonged storage time [62], which was not the case. Simultaneously, the previous blanching of cashew nuts in hot water helps destroy microorganisms such as bacteria, yeasts, and molds [63].

Concerning yeasts, FCNCA-CC and FCNCA-P register < 2 log CFU·g^{-1}, whereas FCNCA-C exceeded 3.0 log units, showing statistical differences for FCNCA-C vs. FCNCA-CC and FCNCA-C vs. FCNCA -P (p = 0.0139). The results indicated that seaweeds could inhibit yeast's activity, which is unsurprising, since some demonstrate anti-fungal capacity against yeast strains [64].

Results of *Escherichia coli* ranged from <1 log CFU·g^{-1} to 2.11 log CFU·g^{-1}, showing statistical differences for FCNCA-C vs. FCNCA-CC and FCNCA-CC vs. FCNCA-P (p = 0.0126). Mendes et al. [65] demonstrate that ethyl acetate and diethyl ester extracts of

C. crispus from the wild and from an integrated multi-trophic aquaculture system possess antimicrobial activity against the growth of bacteria such as *E. coli* and others.

Total coliforms (TCs) ranged from 0.7 to 2.57 log CFU·g^{-1}, revealing differences for FCNCA-C vs. FCNCA-P and FCNCA-CC vs. FCNCA-P (p = 0.0126). The FCNCA-P shows a high content of total coliforms.

Counts of coagulase-positive *S. aureus* reached similar counts between samples, ranging from 2.18 to 2.72 log CFU·g^{-1}. The statistical analyses revealed significant differences for FCNCA-C vs. FCNCA-P (p = 0.0194). The low values for *S. aureus* were considered satisfactory from a safety point of view since the production of toxins only occurs at higher counts (>10^4 or 4 log CFU·g^{-1}) [14].

Salmonella spp. was absent through the plating count (not present per 25 g). In fact, a US surveillance study shows that the prevalence of *Salmonella* (95% of confidence interval) in cashew nuts was minimal (0.55%), i.e., it occurred in just 4 of 733 samples [66].

Listeria monocytogenes was also absent through the plating count (not present per 25 g). Eglezos [67] reported that "there are no data available on the prevalence of *L. monocytogenes* in cooked edible nut kernels or any foodborne illness lined to the presence of *L. monocytogenes* in this kind of product". On the other hand, LAB, which is one of the best represented groups in the FCNCAs (5.48–6.56 log CFU·g^{-1}), has shown bactericidal and bacteriostatic properties against foodborne pathogens such as *Salmonella* spp. and *L. monocytogenes* [19].

3.11. Flash Profile Methodology

Results obtained in the second session of the Flash Profile from the eight panelists ranking for the FCNCAs (FCNCA-C, FCNCA-CC, and FCNCA-P) per attribute, according to the final list of terms defined, were analyzed by Generalized Procrustes Analysis (GPA).

FCNCA-C showed the most consensual rankings, as it presents the lowest residual variance for all attributes, except for texture, for which it presents the highest residual variance (20.6%) (Table 6).

Table 6. Residual variance values for each fermented cashew nut cheese alternatives from Flash Profile's Generalized Procrustes Analysis.

Attributes	Object	Residual (%)
Appearance	FCNCA-C	12.412
	FCNCA-CC	29.920
	FCNCA-P	13.578
Aroma	FCNCA-C	9.729
	FCNCA-CC	11.096
	FCNCA-P	17.230
Flavor	FCNCA-C	6.051
	FCNCA-CC	18.133
	FCNCA-P	13.764
Texture	FCNCA-C	20.604
	FCNCA-CC	16.473
	FCNCA-P	15.811
After-taste	FCNCA-C	7.781
	FCNCA-CC	22.348
	FCNCA	12.737

Fermented cashew nut cheese alternative with *C. crispus* (FCNCA-CC) had the highest residual variances for appearance (29.9%), flavor (18.1%), and after-taste (22.3%). In comparison, FCNCA-P had the highest residual variance for aroma (17.2%), showing that the supplementation of seaweeds, an ingredient that is not very common, influences the sensory characteristics of products and consequently affects the consensus.

Residual variances values for each panelist calculated by GPA reveals the panelists with higher residual variance values: a higher percentage for appearance for panelist 7 (14.1%), aroma for panelist 8 (10.2%), flavor for panelist 1 (15.0%), texture for panelist 4

(22.2%), and after-taste for panelist 1 (13.4%), indicating that these panelists were further from the consensus (see Supplementary Materials, Table S8).

The values of consensus index (Rc) (Table 7) were as follows: appearance (33.8%), aroma (31.2%), flavor (29.1%), texture (11.6%), and after-taste (23.4%). All attributes show an inadequate consensus in the performance of the panelists, particularly on texture and after-taste.

Table 7. Consensus index (Rc (%)) determined among the panelists for each attribute (appearance, aroma, flavor, texture, and after-taste) of the fermented cashew nut cheese alternatives.

Attributes	Rc (%)
Appearance	33.8%
Aroma	31.2%
Flavor	29.1%
Texture	11.6%
After-taste	23.4%

Figure 3 shows the coordinates of the objects (FCNCAs) after GPA analysis and the correlations between the attributes (appearance, aroma, flavor, texture, and after-taste) and the dimensions F1 (first axis) and F2 (second axis).

For the appearance attribute (*a*), the FCNCA-C is perceived as cohesive, porous, and with holes, whereas FCNCA-CC is brownish, greasy, purple, rose/rosy, and soft, and FCNCA-P is opaque. Although there is a discrepancy between the terms used to describe FCNCA-C, the terms porous and with holes are in line with TPA findings, with FCNCA-C showing the lowest values for hardness (6.01 N) and cohesiveness (0.33). FCNCA-CC is described by terms such as purple, rose/rosy, and pink seaweeds, corresponding with its value (3.19) for the color parameter a^*. The opacity related to FCNCA-P concords with results for lightness (L^*) and chroma (C^*), since it shows the lowest values for this parameter, namely, 38.30 and 15.20, respectively.

Regarding aroma (*b*), FCNCA-C was mainly characterized by cheese, dried fruits, *Flamengo* cheese, fermented, and yogurt, whereas FCNCA-CC was characterized by cigar, *nam pla*, salty, straw of coffee, and wood, and FCNCA-P by dry herbs.

Terms such as cheese, *Flamengo* cheese, fermented, or even yogurt suggest a similitude between FCNCA-C and dairy cheese, due to the addition of *L. acidophilus* LA-5®. In fact, FCNCA-C is the sample that shows the higher count of LAB (6.56). The aroma of dried fruits attributed to FCNCA-C is desirable, since dried fruit is at the product's base. Lima et al. (2012) [68] reported an analog term, nutty, to a cashew nut butter made with different grades of kernel quality.

In red seaweeds, the formation of apocarotenoids such as β-ionone contributes significantly for the aroma of algae and marine environments [69], as suggested by terms such seaweeds, shellfish, and sea air. The fishy aroma of *nam pla* may arise from compounds such as 1-octen-3-one, from various aldehydes; for example, when combined, hexanal, (2*E*,4*E*)-decadienal and (2*E*,4*E*)-heptadienal, heptanal can create a strong and penetrating fishy odor, often associated with seaweeds [70]. Furthermore, dry herbs described for FCNCA-P can be associated with a green odor related to esters [71].

The results for flavor (*c*) show that FCNCA-C was mainly characterized by bread and nauseating; FCNCA-CC by sour; and FCNCA-P by Dulse, Nori, and spices.

It can be inferred that some of the panelists, despite being trained, were confused about the flavor of seaweeds, since FCNCA-P was described as Dulse (*Palmaria palmata*) and Nori (*Porphyra* sp.), which is quite understandable, as sometimes aromas and flavors can be very complex and difficult to distinguish from each other. Another term attributed to FCNCA-P, namely, 'spices', is related to carboxylic acids [71].

Regarding texture (*d*), FCNCA-C was characterized mainly by cohesiveness, hardness, and adhesiveness, while FCNCA-CC by terms such as arenaceous, dry, hardness of particles, and light, and FCNCA-P by brittle.

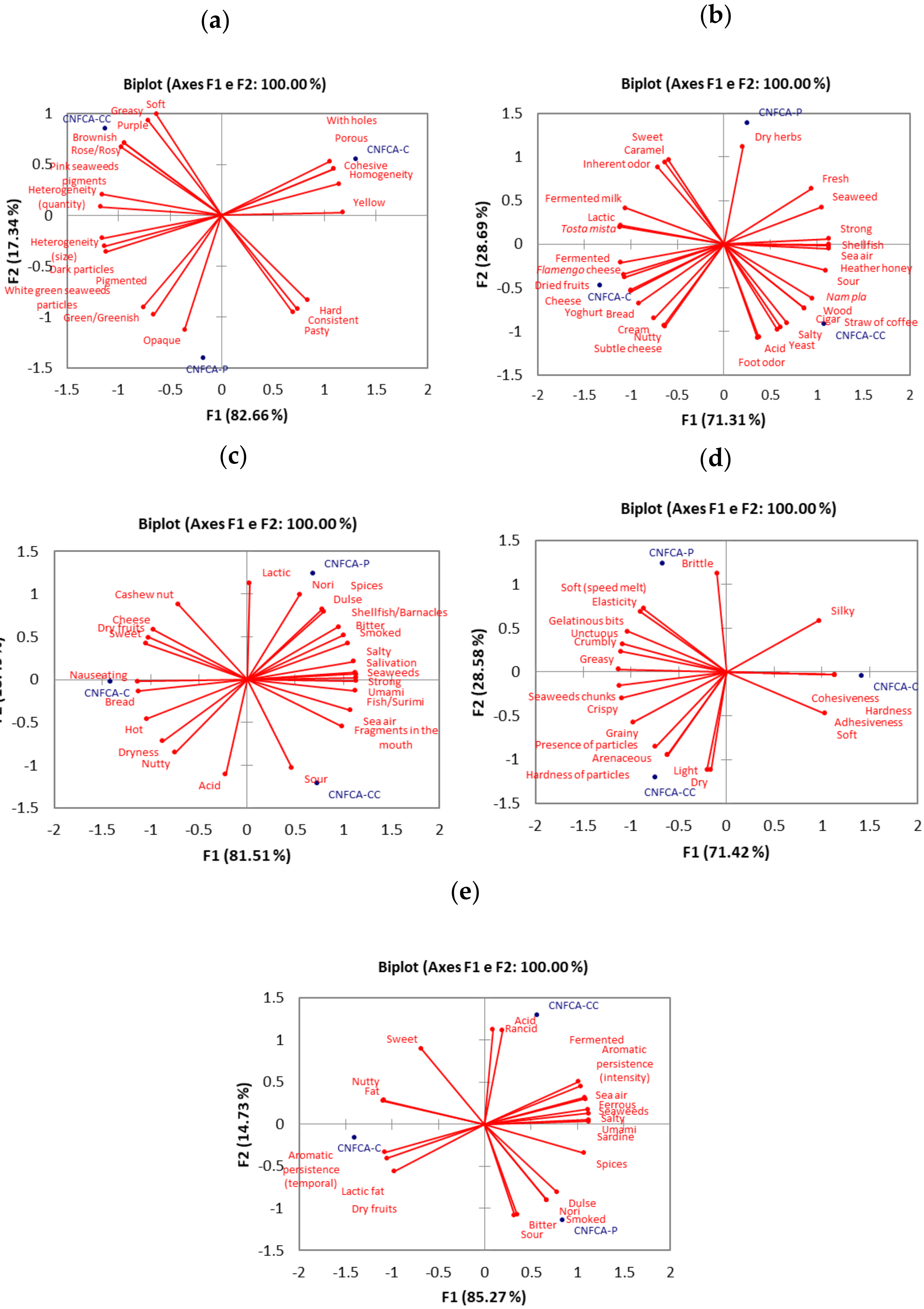

Figure 3. Biplot map of Generalized Procrustes Analysis (GPA) performed on Flash Profile (FP) data and the lexicon used to describe the various attributes of the fermented cashew nut cheese alternatives at the F_1 and F_2 dimensions: (**a**) appearance; (**b**) aroma; (**c**) flavor; (**d**) texture; and (**e**) after-taste.

Dry and brittle terms described for FCNCA-CC and FCNCA-P also comply with the founded hardness values, namely, 7.90 and 9.69 N, respectively. Particle hardness for FCNCA-CC can be attributed to the texture of *C. crispus*, which is generally firm or cartilaginous-like [72]. On the other hand, the size of the particles (2.87 mm) may also have influenced the sensation described.

For after-taste *(e)*, FCNCA-C is characterized by several terms, such as aromatic persistence (temporal), lactic fat, nutty fat, and dry fruits. In contrast, FCNCA-CC is characterized by acidic and rancid, and FCNCA-P by bitter, Dulse, Nori, smoked, and sour.

During the fermentation, lactic acid is produced through sugar metabolism, which is responsible for the increasing sourness as the sweetness reduces [58]. In general, the supplementation of food products with seaweeds compromises their sensory attributes due to the appearance of off-flavors [73], for example, rancidity, as described for FCNCA-CC. This rancidity can be attributed to aldehydes, including heptanal and octanal, which contribute to undesirable rancid odor and flavor during spoilage of fat and fatty foods [74,75]. Also, aldehydes such as heptanal and 2-octenal are associated with 'smoked' and 'sour' [75], respectively, both attributed to FCNCA-P.

In summary, seaweeds have a decisive role in various attributes, especially aroma, flavor, and after-taste, which changed the descriptions of all the attributes, making it possible to distinguish FCNCAs clearly.

4. Conclusions

The cashew nut fermented cheese alternatives can be seen as an interesting alternative to dairy cheese consumption, as they provide an overall nutritionally healthier profile.

Supplementation with seaweeds influenced the technological and sensory characteristics of the FCNCAs and thus contributed to conferring different characteristics to the products, enhancing diversity.

Total lipid and crude protein contents of the supplemented FCNCAs show slightly lower values, revealing that seaweeds do not contribute significantly to these parameters. Macro- and microminerals increased mainly for the FCNCA-CC, namely, Ca, K, Mg, Na, Fe, I, Se, and Zn.

Concerning microbiota, all parameters comply with the European guidelines, except for AMB in all samples and MAC in both supplemented products. Furthermore, the data obtained suggest the presence of a spontaneous lactic fermentation, possibly due to the addition of capsules containing *Lactobacillus acidophilus*, which was responsible for the production of substances that can improve several aspects of products (e.g., flavor, texture, nutrition, shelf-life, and food safety).

The main impact of adding seaweeds to the food matrix was related to the organoleptic characteristics of the developed products. The instrumental color and some of the textural parameters (hardness, springiness, and gumminess) were significantly influenced as well as the sensory characteristics. The sensory profiling of the supplemented products showed their very distinguishable characteristics, thus having high potential to contribute to increase the diversity of products available for consumers that require alternatives to dairy products, as well as allowing the familiarization of consumers with seaweeds.

This was a first exploratory study, and further research is needed to improve the manufacturing process, explore the incorporation of other types of seaweed and their mixtures, and test various concentrations of seaweed. Additionally, evaluating consumer acceptance of these products is also crucial.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/app142311082/s1, Table S1: Methods used for the various microbiological parameters (*Enterococcus*, Lactic Acid Bacteria (LAB), aerobic mesophilic bacteria (AMB), marine agar counts (MACs), Glucose–Yeast–Peptone (GYP) molds and yeasts, *Escherichia coli*, total coliforms (TCs), *Staphylococcus aureus*, *Salmonella* spp., and *Listeria monocytogenes*); Table S2: Mann–Whitney U test pairwise comparisons between fermented cashew nut cheese alternatives (FCNCAs): fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. FCNCA-P) for general physicochemical parameters; Table S3: Mann–Whitney U test pairwise comparisons between fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) for minerals and trace elements; Table S4: Mann–Whitney U test pairwise comparisons between fermented cashew nut cheese alternatives (FCNCAs): fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) for color parameters; Table S5: Mann–Whitney U test pairwise comparisons between fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) for TPA parameters (hardness (N), adhesiveness (J), cohesiveness (—), springiness (mm), and gumminess (N)); Table S6: Mann–Whitney U test pairwise comparisons between fermented cashew nut cheese alternatives: fermented cashew nut cheese alternative control (FCNCA-C); fermented cashew nut cheese alternative with *Chondrus crispus* (FCNCA-CC); and fermented cashew nut cheese alternative with *Porphyra* sp. (FCNCA-P) on microbiological parameters (*Enterococcus*, Lactic Acid Bacteria (LAB), aerobic mesophilic bacteria (AMB), marine agar counts (MACs), Glucose–Yeast–Peptone (GYP) molds and yeasts, *Escherichia coli*, total coliforms (TCs), *Staphylococcus aureus*, *Salmonella* spp., and *Listeria monocytogenes*); Table S7: Guidance criteria on the interpretation of microbiological results in ready-to-eat foods placed on the market (*Enterococcus*, Lactic Acid Bacteria (LAB), aerobic mesophilic bacteria (AMB), marine agar counts (MACs), Glucose–Yeast–Peptone (GYP) molds and yeasts, *Escherichia coli*, total coliforms (TCs), *Staphylococcus aureus*, *Salmonella* spp., and *Listeria monocytogenes*); Table S8: Residual variance values, scaling factors, and the percentage variation explained by the first two principal components (F1 and F2) of Generalized Procrustes Analysis (GPA) for fermented cashew nut cheese alternative (FCNCA) Flash Profile analysis.

Author Contributions: Conceptualization, P.M., M.S.D. and B.M.C.; methodology, P.M., M.S.D. and B.M.C.; software, M.S.D. and B.M.-L.; validation, P.M. and M.S.D.; formal analysis, B.M.C., B.M.-L., A.S., E.R., I.M., M.M.-F. and P.S.; investigation, B.M.C.; data curation, P.M., M.S.D., M.M.-F. and P.S.; writing—original draft preparation, B.M.C.; writing—review and editing, B.M.C., P.M. and M.S.D.; supervision, P.M. and M.S.D.; funding acquisition, P.M., M.S.D. and B.M.-L. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the European Maritime and Fisheries Fund and co-funded by the Operational Program Mar2020 through the project Alga4Food (MAR-01.03.01-FEAMP-0016). This research was also supported by the Applied Molecular Biosciences Unit (UCIBIO) and the Associate Laboratory for Green Chemistry (LAQV), both funded by national funds from FCT/MCTES (10.54499/UIDB/04378/2020) and (10.54499/UID/50006/2020), respectively. This study was also co-financed by the ERDF under the PT2020 Partnership Agreement (POCI-01-0145-FEDER-007265).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Data are contained within the article and Supplementary Materials.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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