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The Biotransformation of Lupine Seeds by Lactic Acid Bacteria and *Penicillium camemberti* into a Plant-Based Camembert Alternative, and Its Physicochemical Changes during 7 Weeks of Ripening

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Abstract: In recent years, there has been increasing consumer interest and research into plant-based dairy alternatives, due to the increasingly negative impact of animal products on human health, animal welfare, and the environment. The purpose of this study was to investigate the physicochemical and microbiological changes in a Camembert alternative based on the seeds of sweet lupine (*Lupinus angustifolius* L cv. 'Boregine'). After heat treatment and homogenization, the seeds were incubated with lactic acid bacteria (LAB) and *Penicillium camemberti* mold. After fermentation at room temperature, the samples were stored at 12 °C for 14 days, and then ripened until day 49 at 6 °C. Changes in microbial population, acidity, texture, content of polyphenols, flavonoids, reducing sugars, and free amino acids were monitored. In addition, the antioxidant capacity of the samples during ripening was determined. The results showed that LAB and fungi were able to grow well in the lupine matrix. Initially, a decrease in pH was observed, while in the further stages of ripening, alkalization of the product linked with progressive proteolysis associated with an increase in free amino acid content was noted. Hydrolysis of polysaccharides and an increase in antioxidant activity were observed. This indicates the potential of lupine seeds as a raw material for the development of a new group of plant-based ripened cheese alternatives.

Keywords: cheese alternatives; ripening; lupine; plant-based foods

Citation: Łopusiewcz, Ł.; Śmietana, N.; Lichwiarska, E.; Mazurkiewicz-Zapałowicz, K.; Gefrom, A.; Drozłowska, E. The Biotransformation of Lupine Seeds by Lactic Acid Bacteria and *Penicillium camemberti* into a Plant-Based Camembert Alternative, and Its Physicochemical Changes during 7 Weeks of Ripening. *Fermentation* **2022**, *8*, 447. https://doi.org/10.3390/fermentation8090447

Academic Editor: Michela Verni

Received: 22 August 2022
Accepted: 4 September 2022
Published: 8 September 2022

1. Introduction

Milk and milk products are considered a class of foods containing compounds essential to human nutrition [1,2]. Dairy remains an important and major agricultural product, including but not limited to fermented foods such as yogurt, kefir, and cheese [3]. Cheese is classified as a dairy product, easily digestible with high biological and nutritional value [4]. However, a plant-based lifestyle is becoming increasingly popular, and people are switching from animal-based diets to plant-based diets in recent times due to health, ethical, environmental, and socioeconomic concerns [1–3,5–7]. In addition, recent interest in new foods and beverages is driving the production of partially substituted and plant-based dairy analogs and alternatives [8]. In 2020, the COVID-19 pandemic accelerated this process, as it forced consumers to rethink their lifestyles and switch to a more plant-based diet as a healthier option [7]. Therefore, food researchers are working to develop new food products that meet the broad expectations of consumers [6]. Plant-based cheese

substitutes are defined as products prepared in whole or in part using plant-based ingredients (non-dairy fats and proteins). These products do not contain cholesterol but have more fiber and complex carbohydrates [8]. These plant-based dairy alternatives are likely to be more environmentally sustainable and better for animal welfare than their regular counterparts [2]. Plant-based cheeses involve consolidating the protein mass from various plant sources with molds, yeast, and lactic acid bacteria that may also be added for acidity. "Cheese-like" products made from soy (such as tofu) or nuts are among the most popular vegan products [9–13]. The market for cheese-like plant-based products is still growing. It is assumed that it could expand at a CAGR of 12.5% and reach a global market size of 52.58 billion USD by 2028 [8]. The growing popularity of this market has resulted in an expanding range of dairy alternatives involving a huge variety of plant matrices [3,5,14–16]. Therefore, plant-based cheeses are a promising future food that has the potential to serve as a nutritious and sustainable alternative to regular cheese [2].

During cheese ripening, biochemical changes can be divided into primary events, such as proteolysis, lipolysis, and glycolysis, and secondary events, such as fatty acids and amino acids metabolism [17,18]. They are both pivotal for cheese flavor and aroma development [17,19–21]. They result from the action of enzymes either produced by live microorganisms or released onto the cheese matrix after microbial cellular lysis [5]. Camembert is one of the most well-known ripened cheeses and is characterized by a velvety white layer originated by the growth of fungi (mold *Penicillium camemberti* and optionally yeast *Geotrichum candidum*). Camembert-type cheeses ripen quickly due to a high moisture content and rapid growth of surface molds [19–21]. The ripening of Camembert is very complex, and intense proteolytic activity (mainly from microbial origin) gives this kind of cheese its specific texture, flavor, characteristic aroma, and bioactivity. Several plant matrices are reported to have been used for ripened cheese-like products, such as flaxseed oil cake, peanut, cashew, pea protein isolate, rice flour, and apricot pulp [5,22–24]. However, studies of bioactivity and physicochemical properties during the ripening of plant-based Camembert alternatives are still very limited.

Lupine belongs to the family of *Fabaceae*, also known as legumes, that is rich in protein (up to 44%) and phenolic compounds (such as flavonoids and polyphenols), phytosterols, and squalene [2,12,25]. It is also important that lupine is seen as a valuable source of nutrients. In addition to protein, lipids, dietary fiber, minerals, and vitamins are also present in lupine seeds [13,26]. Phytochemicals from lupine seed exhibit an antioxidant capacity [27]. The plant has many varieties, as the *Lupinus* genus includes more than four hundred species. It should also be mentioned that four of them are represented as sweet lupine [28]. Lupine cultivation is an important part of the agricultural sector in Europe. The current lupine market in Europe reached 284 million in 2019 and continues to grow. It is expected to become profitable by 2027, growing at a rate of 4.39% over the forecast period 2020–2027. The potential use of whole seeds and parts of the plant is widely reported in the literature [12,29–33]. Moreover, lupine can be successfully used a raw material in the production of dairy substitutes [3,34–36]. However, its application as a matrix for the development of a plant-based Camembert alternative using lactic acid bacteria and mold *Penicillium camemberti* has not been reported so far.

The purpose of the present work was to analyze the possibility of obtaining a Camembert alternative using lupine seeds and to evaluate its bioactivity, microbiological, and physicochemical properties during 49 days of ripening.

2. Materials and Methods

2.1. Materials and Chemicals

Lupine seeds (*Lupinus angustifolius* L. cv. "Boregine"—with low alkaloid content < 0.01%) were kindly donated by Saatzucht Steinach GmbH & Co KG (Steinach, Germany). Commercial starter cultures: PC® (containing *Penicillium camemberti*) and MST Cheese-Tek® (containing *Lactococcus lactis su*bsp. lactis, *Lactococcus lactis subsp. cremoris* and *Streptococcus salivarius subsp. thermophilus*) were supplied by Biochem s.r.l. (Biochem s.r.l, Rome, Italy). Sodium hydroxide,

disodium phosphate, monosodium phosphate, 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), methanol, Folin-Ciocalteu reagent, sodium chloride, sodium carbonate, gallic acid, 3,5-dinitrosalicylic acid, sodium tartrate tetrahydrate, acetic acid, sodium acetate, potassium ferricyanide, trichloroacetic acid, ferric chloride, ninhydrin, glacial acetic acid, cadmium chloride, glycine, ferric chloride hexahydrate, and 2,4,6-tripyridyl-s-triazine (TPTZ), quercetin were purchased from Merck (Merck, Darmstadt, Germany). Glucose, hydrochloric acid, sodium nitrite, aluminum chloride, and ammonium thiocyanate were supplied from Chempur (Chempur, Piekary Śląskie, Poland). All reagents were of analytical grade. Microbial analyses were conducted with MRS agar, and Sabouraud agar supplemented with chloramphenicol (0.05%) obtained from Merck (Merck, Darmstadt, Germany).

2.2. Products Preparation, Fermentation, Ripening, Microbial Analyses, and Acidity Determination

The lupine matrix was prepared in several steps. Lupine seeds were washed with distilled water (*w/w*) and stored in the refrigerator (6 °C) for 24 h. The drained seeds were mixed with distilled water (1400 g of seeds per 3000 mL of water), the mixtures were heated at 90 °C for 20 min with constant stirring (250 rpm), and cooled to room temperature. The samples were homogenized for 5 min with a homogenizer (SilentCrusherM, Heldolph, Germany) at 12,000 rpm and the mixture was pasteurized (60 °C, 30 min). After homogenization, the sample was inoculated in sterile conditions (laminar flow cabinet Polon KLVS-1, Poznań, Poland) with 0.5 g of MST Cheese-Tek® and 0.5 g of PC®. The mixture was additionally homogenized with a domestic mixer, and dispensed into sterile plastic Camembert cylindrical forms (Serowar, Szczecin, Poland) layered down with a gauze (200 ± 1 g of mixture per one form). The fermentation was conducted at room temperature (25 ± 1 °C) for 24 h. After this time, the samples were removed from the forms and the gauze was slightly removed, then salted with 1.5 g of NaCl per sample. The ripening process was carried out in a climatic chamber (Binder FD15, Tuttlingen, Germany) on cheese maturation mats at 12 °C for 14 days. The samples were turned daily, and on day 14 were wrapped in polyethylene-covered paper (0.06 mm thickness, Serowar, Szczecin, Poland); the conditions were changed (6 ± 1 °C, 90% RH), and the samples were ripened until day 49.

The microbial analyses were performed during the overall storage. Samples (10 g) were collected and diluted with 90 mL of sterile physiological saline (0.9% NaCl), and serial dilutions were prepared [5]. Lactic acid bacteria (LAB) counts were determined on MRS (de Man, Rogosa and Sharpe) medium (Merck, Darmstad, Germany) after incubation at 37 °C under anaerobic conditions for 48 h. The fungal counts were marked on Sabuorad Agar with Chloramphenicol at 25 °C for 72 h. Each enumeration of microorganisms was performed in triplicate. Final outcomes of the level cell viability were expressed as CFU/g of the samples.

The pH was measured in prepared saline after homogenization with samples directly at 25 °C using a pH-meter (CP-411, Elmetron, Zabrze, Poland).

2.3. Determination of Bioactive Compounds

Cheese-like products were lyophilized for 24 h (chamber pressure 0.190 mbar, shelf temperature T_{min} = −35 °C, T_{max} = 20 °C, condenser temperature −85 °C) in Beta 2–8 LSC plus lyophilizer (Martin Christ Gefriertrocknungsanlagen GmbH, Osterode am Harz, Germany). The extracts were prepared as previously described [5,29]. The obtained clear fluids were used for further analyses. The Total Polyphenols Content (TPC) and Total Flavonoids Content (TFC) were analyzed spectrophotometrically using a microplate reader (Synergy LX, BioTek, Winooski, VT, USA) by placing the samples in a 96-well microplate. TPC was measured with by the Folin–Ciocalteu method described by Tong et al. [28]. An amount of 100 µL of the extract was mixed with 6 mL of distilled water and 0.5 mL of Folin-Ciocalteu reagent. For the previous 3 min of incubated mixture, 1.5 mL of saturated

Na_2CO_3 solution was added. Next, the samples were incubated for 30 min in the dark at 40 °C. Finally, the absorbance was measured at 765 nm and the outcomes expressed as mg gallic acid equivalents (GAE) per mL of sample (mg GAE/mL).

TPC evaluation was based on the method described elsewhere [5]. With the aim of conducting the analysis, 250 µL of the sample were mixed with 1 mL of distilled water and 75 µL of 5% $NaNO_2$ solution. After 5 min incubation, 75 µL of 10% $AlCl_3$ solution was added into mixture and the whole sample was allowed to stand for 6 min before the addition of 250 µL of 1 M NaOH. Finally, the total volume mixture was made up to 3 mL with distilled water, and then the absorbance was measured at 510 nm. Quercetin was used for a calibration curve, and the results were expressed as mg of quercetin equivalents (QE) per g of the sample (mg QE/g).

The Reducing Sugar Content (RSC) was measured with the DNS (3,5-dinitrosalicylic acid) method as described elsewhere [5]. The extracts were mixed with 1 mL of 0.05 M acetate buffer (pH 4.8) and 3 mL of DNS reagent and shaken vigorously. Then, samples were incubated in boiling water for 5 min and cooled at room temperature. The absorbance value was measured at 540 nm using a microplate reader. Glucose (0.01–10 mg/mL) in acetate buffer was used for the calibration curve.

Total free amino acids (TFAA) were measured using Cd-ninhydrin reagent [5]. The extracts were mixed with reagent and heated at 84 °C for 5 min, cooled in ice water, and the absorbance at 507 nm was measured. The results were expressed as mg glycine (Gly) per gram of sample with respect to the standard curve including the dilution factor. The standard curve was first prepared using glycine (0.01–10 mg/mL).

2.4. Determination of Antioxidant Activity

The DPPH, ABTS, and FRAP radical scavenging activity were determined according to the procedures as described in a previous study [5,29]. Additionally, the Reducing Power (RP) test was chosen. In order to determine the DPPH radical scavenging activity, one milliliter of the methanolic supernatants was mixed with 1 mL of 0.01 mM DPPH methanolic solution. The absorbance was measured at 517 nm. The $ABTS^{\cdot+}$ solution was mixed with 50 µL of the methanolic supernatants. In this test, absorbance was measured at 734 nm.

The FRAP scavenging activity was performed by mixing 25 mL of acetate buffer (300 mM), 2.5 mL of 2,4,6-tripyridyl-s-triazine (TPTZ) solution (10 mM in 40 mM HCl), and 2.5 mL of ferric chloride hexahydrate aqueous solution (20 mM). To 300 µL of FRAP reagent in a microcentrifuge tube, 10 µL of extracts were added and vortexed for 10 s. Absorbance was measured at 593 nm [37].

To determine RP, the samples (500 µL) were placed in tubes to which 1.25 mL of phosphate buffer solution (0.2 M, pH 6.6) and 1.25 mL of 1% potassium ferricyanide solution were added, and incubated at 50 °C for 20 min. Next, 1.25 mL of trichloroacetic acid solution was added to the tubes. The supernatant obtained by centrifugation at 3000 rpm for 10 min was diluted 1:1 with of deionized water. In the final step, 0.25 mL of 0.1% ferric chloride solution was added to complete the determination of the reduction of ferric ion (Fe^{3+}). The measurements were carried out at 700 nm.

2.5. Textural Analyses

Texture profiles of the samples were performed at room temperature using a Zwick/Roell 2,5 Z equipment (Zwick/Roell, Ulm, Germany), equipped with a cylindrical probe (diameter 40 mm). The samples were analyzed on each research day, and the penetration rate into the samples was 10 mm/s, while the depth was 25 mm. From the results of the force-time curves, the hardness, gumminess, springiness, chewiness, and cohesiveness were calculated.

2.6. Statistical Analysis

The statistical analyses were conducted using Statistica version 13 (StatSoft Polska, Kraków, Poland). In order to obtain statistically important results, all experiments were replicated three times. All the outcomes have been expressed as mean ± standard deviation (SD). Two-way variance analyses (two-way ANOVA) were followed by Fisher's NIR test. Values were considered as significantly different when $p < 0.05$.

3. Results and Discussion

3.1. The Changes in Microbial Population, Acidity, and Reducing Sugars Content during the Ripening

In the context of fermented foods, biotransformation by fermentation refers to a general process in which a food matrix is metabolized/transformed by microorganisms to impart desirable properties [3]. As shown in Figure 1, the LAB and fungal survivability was high during the ripening process. The lupine-based formulation provided an excellent matrix for the development of a new plant-based Camembert alternative. The initial content of LAB was $9.75 \times 10^7 \pm 0.21$ CFU/g, whereas the fungi content was $1.11 \times 10^6 \pm 0.01$ CFU/g. During the ripening, these values significantly increased ($p < 0.05$). The LAB content increased from 0 to 7 day ($p < 0.05$). The highest LAB value was observed on day 35 ($3.16 \times 10^{12} \pm 0.05$ CFU/g). Those results are comparable with the observations of Lee and Bae for milk Camembert [38], as well as for cashew cheese analogs [39]. A similar LAB content was described by Bartikiene et al., in the case of lupine flour in the semisolid fermentation process [40]. However, the obtained values are higher than those obtained in the previous study where a flaxseed press cake was used as a raw material for a Camembert alternative production [5]. Furthermore, the results were also similar to LAB counts obtained in the lupine milk fermentation process described by Jimenez-Martinez [35]. The highest fungal counts were observed on day 21 ($3.18 \times 10^8 \pm 1.06$ CFU/g), after which some decrease in fungal viability was noticed ($p < 0.05$). Lupine could be successfully fermented by fungi.

According to Trojanowska et al. [41], lupine seeds contain various oligosaccharides, which could be metabolized as substrates for growth. Moreover, the formation of a white, velvety crust was observed during the ripening period (Figure 2). A similar observation was reported in a previous study using *P. camemberti* for biotransformation of flaxseed oil cake [5]. In fact, *P. camemberti* is an aerobe and always grows on the surface of cheese [42].

As shown in Figure 3, a significant reduction in pH was observed from day 0 to day 14, after which the pH of the samples significantly increased during the ripening period ($p < 0.05$). The reduction in pH is linked with the production of organic acids (mainly lactic acid) by LAB, as in the acidification of cheese milk, which is their well-known function in cheese production [5]. In fact, a similar reduction in pH was reported for a Camembert alternative produced from flaxseed oil cake [5]. The increase in pH after day 14 can be attributed to fungal activity, and medium alkalization resulting from the assimilation of organic acids, decarboxylation, and deamination of amino acids, as well as production of ammonia [21,38]. The highest pH (8.35 ± 0.10) was observed on day 49 and is higher than reported for sufu [4], for a Camembert alternative made from flaxseed oil cake [5], as well as milk Camembert [38].

The changes in RSC are presented in Figure 4. As can be seen, a significant ($p < 0.05$) increase in RSC content was observed after day 3. The highest RSC content was observed on day 14 (74.70 ± 1.72 mg/g), and after that it decreased sharply ($p < 0.05$). Similar observations were reported in a previous study using flaxseed oil cake [5], however the RSC content in the present study was higher. Obtained results are also in line with the findings of Fritsch et al. [43] who fermented lupine using *Bifidobacterium lactis*. Bacteria and fungi can produce several different types of enzymes, especially hydrolases, which are responsible for breaking down oligosaccharides into simple sugars [5,44]. Thus, it can be concluded that the microorganisms decomposed polysaccharides contained in the lupine matrix, subsequently using the resulting oligosaccharides and simple sugars as substrates

for growth and metabolism. This mechanism is also responsible for the softening of plant material as a result of cell wall degradation, which is of great importance from a sensory, physicochemical, and dietary point of view, since carbohydrate digestibility is linked to many human health problems [5,45].

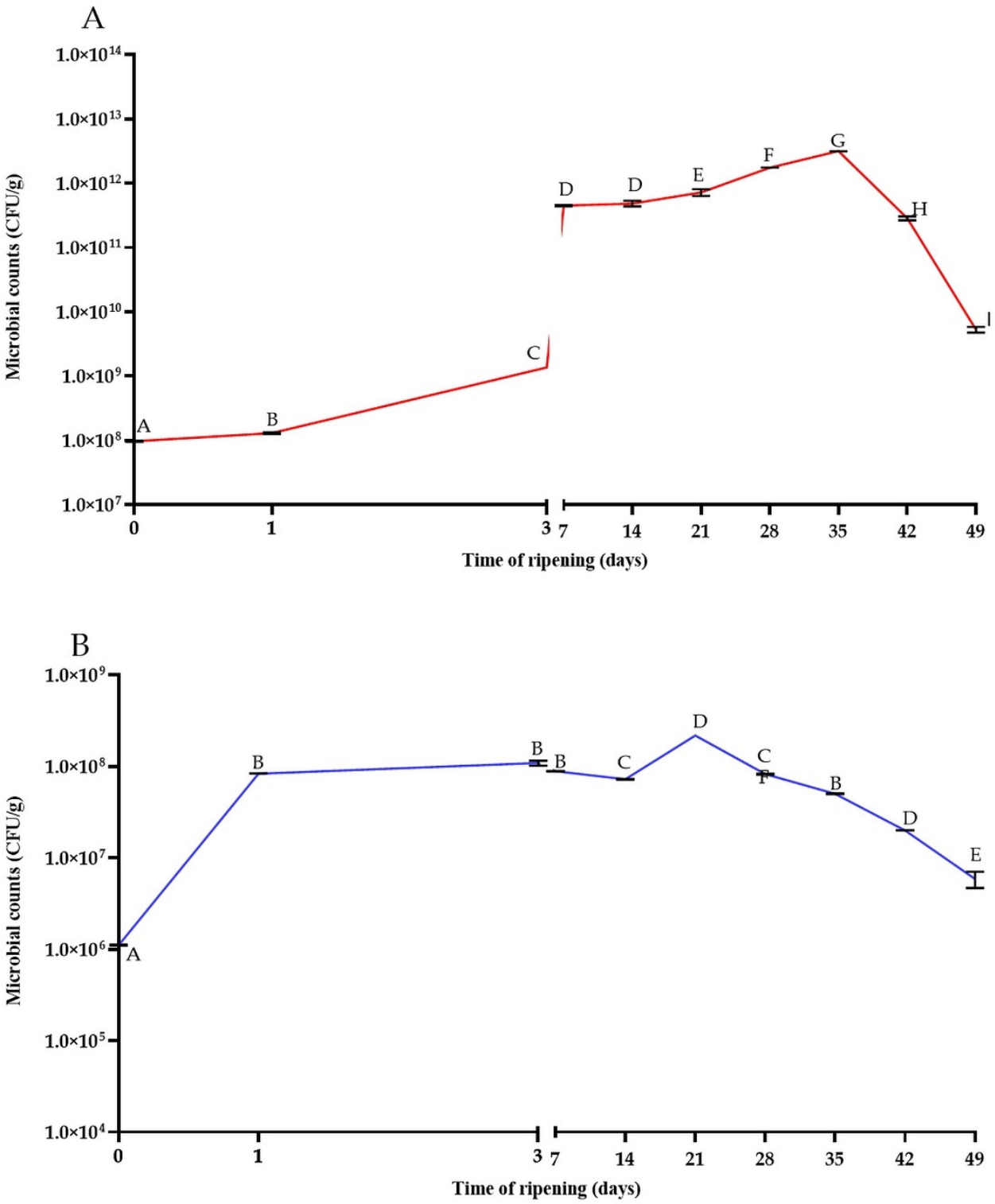

Figure 1. The lactic acid bacteria (**A**) and yeasts (**B**) viability during ripening. Means with different letters (A–I) are significantly different at $p < 0.05$.

Figure 2. A representative photograph of lupine-based Camembert alternative on day 28.

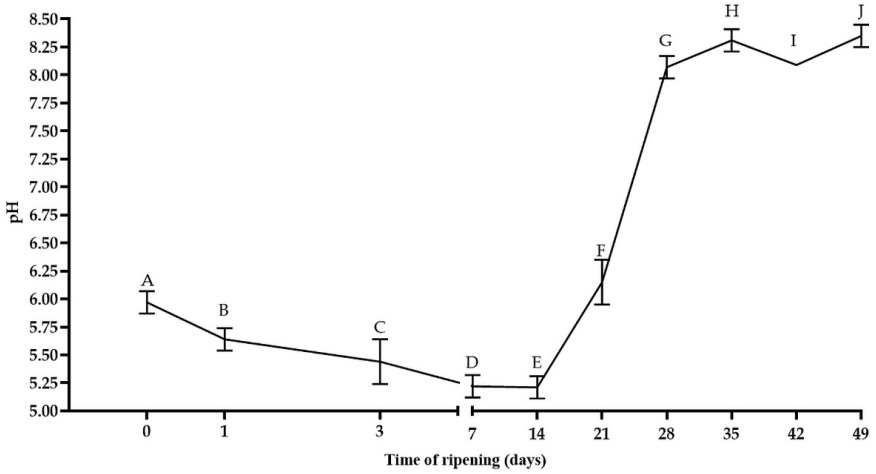

Figure 3. Changes in pH during the ripening time. Means with different letters (A–J) are significantly different at $p < 0.05$.

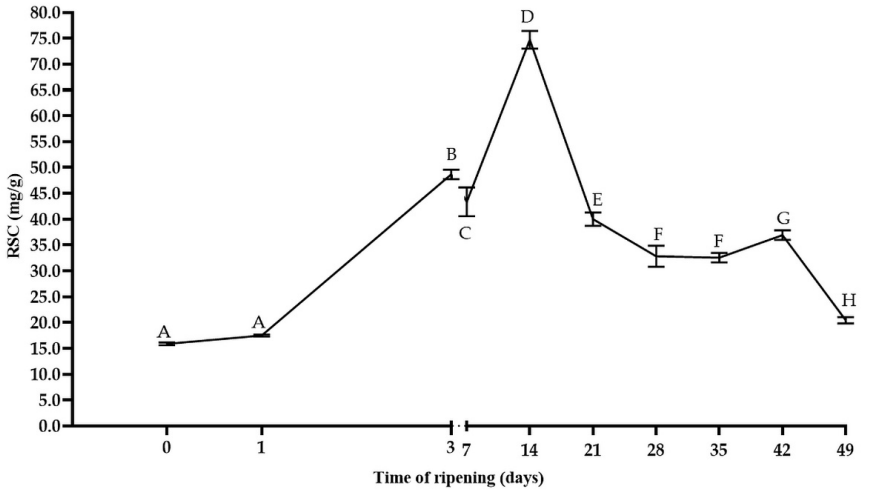

Figure 4. Changes in the Reducing Sugars Content during the ripening. Means with different letters (A–H) are significantly different at $p < 0.05$.

3.2. The Changes in Total Polyphenolics Content, Total Flavonoids Content, Total Free Amino Acids Level, and Antioxidant Activity

The changes in bioactive compounds are presented in Table 1. It is known that fermentation increases the antiradical/antioxidant power as a consequence of reducing metabolites production [46]. In the present study, an increase in TPC was also observed during cheese ripening ($p < 0.05$). There was a more than 7-fold increase in TPC on day 35 (3.46 ± 0.01 mg GAE/g) compared to the initial value (0.52 ± 0.06 mg GAE/g). Moreover, the lupine-based Camembert alternative was more abundant in flavonoids than polyphenols. The initial TFC value was 10.59 ± 0.34 mg QE/g and increased to the highest value of 16.62 ± 0.25 mg QE/g obtained on day 14. The differences between day 0 and day 1 were not statistically significant ($p > 0.05$), but after this time the increase in TFC at a statistically significant level compared to the early stage of the experiment ($p < 0.05$) was observed. Lupine seeds are abundant in bioactive compounds with antioxidant potential, such as polyphenols, mainly tannins, and flavonoids [27]. From a nutritional point of view, the increase in TPC is very important, as these biomolecules are able to prevent chronic diseases such as cancer, diabetes, as well as neurodegenerative and cardiovascular diseases [47]. Polyphenols are also commonly used as anti-allergic, anti-arteriogenic, anti-inflammatory, antimicrobial, antioxidant, and anticoagulant compounds. The fermentation process for legume seeds has a major impact on TPC content. This is due to biomodification between soluble phenolic compounds and the release of bound phenolic compounds by microorganisms involved in the fermentation process [48]. It should also be noted that during fermentation, microorganisms produce phenolic compounds as secondary metabolites. According to Bartkiene et al. [16] high TPC content can be obtained when the matrix is based on whole lupine meal, due to its high dietary fiber content. The increase in TPC is also associated with acidification caused by microorganisms. This process increases the solubility and extractability of polyphenols, which is observed as higher TPC during ripening. It should be noted that this is mainly dependent on the specifics of the LAB and fungal strain and their enzymatic activity. It caused the release of polyphenols from glycosylated and more complex forms with lower activity.

Table 1. The Changes in Total Polyphenolics Content, Total Flavonoids Content, Total Free Amino Acids Level, and Antioxidant Activity.

Time of Ripening (Days)									
0	**1**	**3**	**7**	**14**	**21**	**28**	**35**	**42**	**48**
TPC (mg GAE/g)									
0.52 ± 0.06 ^{A}	0.48 ± 0.05 ^{A}	0.63 ± 0.11 ^{A}	2.15 ± 0.08 ^{AB}	2.32 ± 0.17 ^{B}	2.75 ± 0.27 ^{BC}	2.48 ± 0.26 ^{B}	3.46 ± 0.00 ^{C}	3.00 ± 0.26 ^{C}	2.70 ± 0.30 ^{BC}
TFC (mg QE/g)									
10.59 ± 0.34 ^{A}	10.23 ± 0.46 ^{A}	16.03 ± 0.44 ^{BCD}	16.55 ± 1.13 ^{BD}	16.62 ± 0.25 ^{BD}	15.74 ± 0.13 ^{BCD}	15.37 ± 0.44 ^{C}	15.08 ± 0.46 ^{C}	15.89 ± 0.13 ^{BCD}	15.37 ± 0.76 ^{C}
TFAA (mg Gly/g)									
8.32 ± 0.68 ^{A}	16.68 ± 0.72 ^{A}	45.02 ± 1.20 ^{B}	237.34 ± 2.72 ^{C}	262.21 ± 7.55 ^{D}	276.05 ± 2.30 ^{E}	323.50 ± 2.43 ^{F}	365.50 ± 3.45 ^{G}	245.33 ± 13.49 ^{C}	358.29 ± 4.03 ^{G}
DPPH (%)									
23.62 ± 5.22 ^{A}	34.32 ± 7.34 ^{B}	26.71 ± 2.72 ^{A}	46.06 ± 2.08 ^{CD}	46.19 ± 2.95 ^{CD}	51.51 ± 1.08 ^{D}	35.83 ± 1.61 ^{B}	58.01 ± 2.05 ^{E}	47.24 ± 1.77 ^{CD}	42.72 ± 1.75 ^{C}
ABTS%									
0.46 ± 2.64 ^{A}	1.98 ± 4.12 ^{A}	2.80 ± 1.19 ^{A}	12.57 ± 3.53 ^{B}	17.20 ± 4.43 ^{BC}	20.05 ± 2.14 ^{C}	16.03 ± 2.37 ^{BC}	19.08 ± 2.12 ^{C}	16.23 ± 3.93 ^{BC}	13.89 ± 1.88 ^{B}
FRAP (mg AAC/g)									
0.20 ± 0.01 ^{A}	0.19 ± 0.01 ^{A}	0.22 ± 0.03 ^{A}	0.59 ± 0.02 ^{A}	0.63 ± 0.04 ^{AB}	0.74 ± 0.06 ^{B}	0.67 ± 0.06 ^{B}	0.91 ± 0.00 ^{C}	0.80 ± 0.06 ^{C}	0.73 ± 0.07 ^{BC}
RP (700 nm)									
0.173 ± 0.003 ^{A}	0.187 ± 0.003 ^{AB}	0.214 ± 0.004 ^{B}	0.324 ± 0.007 ^{C}	0.363 ± 0.001 ^{DE}	0.446 ± 0.002 ^{G}	0.307 ± 0.058 ^{C}	0.265 ± 0.021 ^{F}	0.376 ± 0.004 ^{D}	0.340 ± 0.004 ^{CE}

Means with different letters (A–G) in the same raw are significantly different at $p < 0.05$.

The lupine protein is known to be easily fermentable by microorganisms [49]. According to Kasprowicz-Potocka et al. [50], the fermentation process may have caused changes in protein and amino acid content, but strongly reduced true protein content. In fact, it was found that free amino acids concentration was affected by ripening time, which indicates directly progressive proteolysis ($p < 0.05$). The highest TFAAL was noticed on day 49 (358.29 ± 4.03 mg Gly/g), which was approximately 43-fold higher than on day

0 (8.32 ± 0.68 mg Gly/g). A similar observation was reported for classic milk Camembert [19,38], sufu [4], plant-based Camembert alternative produced from FOC [5], as well as *Monascus*-ripened cheese [51]. On the other hand, Zhang et al., reported that liberated amino acids can be consumed to some extent by microorganisms to maintain their metabolic activity [19]. The value on day 49 is higher than for flaxseed-based Camembert [5], although lower than for milk Camembert reported by Lee and Bae [38]. Moreover, an intensive proteolysis can be attributed to pH changes starting on day 14. The proteinases of *P. camemberti* are activated by the increasing pH, and they migrate slowly into the cheese matrix [52]. Proteolysis plays an important role in the texture and flavor of cheeses during ripening, and therefore the degree of proteolysis is considered as an important indicator of cheese maturation [19]. The liberation of free amino acids contributes directly to the flavor of cheeses; moreover, amino acids serve as precursor compounds of the catabolic reactions that generate a number of flavor compounds, such as aldehydes, alcohols, carboxylic acids, thiols, etc. [19].

Table 1 shows the results of radical scavenging activity (DPPH, ABTS, and FRAP) and reducing power (RP). DPPH and FRAP activities significantly increased during the ripening process ($p < 0.05$). Lee and Bae also observed an increase in antioxidant activity in milk Camembert during ripening [38]. The highest values of these scavenging activities were observed on day 35. These results were comparable to those for TFAA and TPC, as the levels of these parameters were also highest on the same day during ripening. It is well known that an increase in total phenols [16], and TFAA, is associated with an improvement in the antioxidant activity of the plant matrix [53]. The increase in ABTS occurred more rapidly, with the highest value observed on day 21 (20.05 ± 2.14%). In the case of RP, the highest level was reached on day 42 (0.376 ± 0.004). In each case, the final results were significantly higher than for the non-biotransformed control sample ($p < 0.05$). The prevention of oxidative stress is nowadays a very important factor. As mentioned earlier, bioactive compounds in lupine seeds can be useful in the treatment of civilization diseases. Free radical scavenging activity may be part of the important processes responsible for scavenging oxidative free radicals and repairing DNA damage. Consumption of antioxidant-rich foods may play an important role in the prevention of many civilization diseases [34,54].

3.3. The Textural Changes

Ripening processes strongly affect the textural characteristics of cheeses [38,51]. In the case of classic milk-based Camembert, the formation of a creamy and uniform appearance occurs, resulting from the degradation and disruption of the casein network structure during the cheese ripening process [5,51,55,56]. As can be seen in Table 2, the texture of the lupine-based product changed significantly during ripening. The hardness of the product tended to increase in the first 2 weeks, and thereafter continued to decrease significantly ($p < 0.05$). A similar trend was reported by Xia et al. for *Monascus*-ripened cheese [51]. It should also be noted that firmness, chewiness, cohesiveness, and gumminess decreased after the fermentation process on the first day ($p < 0.05$). On day 3, cohesiveness and firmness were higher than on the initial day, but the other parameters returned to the initial level ($p > 0.05$). Finally, each parameter was significantly different ($p < 0.05$) and higher except for cohesiveness. Similar observations were reported for a yogurt drink based on lupine powder by Abdel-Salam [14]. These authors also noted that cohesiveness, chewiness, and elasticity decreased after fermentation, while an increase in cohesiveness, gumminess, chewiness, and elasticity was noted during refrigerated storage. Similar trends were also observed in previous studies for Camembert alternatives based on flaxseed cake—a significant increase in hardness, gumminess, elasticity, chewiness, and a decrease in cohesiveness during storage was reported [5]. In milk Camembert, *P. camemberti* metabolizes lactate and produces ammonia through proteolysis, generating a pH gradient from the surface to the core of Camembert cheese, which is responsible for softening the cheese, and changes in the pH of the cheese significantly affect the texture of the final product [38]. It should be

also emphasized that one of the key factors in protein-rich products is the gelation ability of ability of proteins. Lupine protein is well known for its gelling properties and ability to stabilize emulsions. The high amount of protein in a product may be one of the factors that affect texture, especially firmness. In this context, the changes in the texture of the cheese during ripening can be explained by the breakdown of the protein bonds, manifested by the increased amino acids level due to proteolysis [5,57,58]. In the case of textural changes in products of plant origin, it is also necessary to mention the subsequent changes observed during the ripening period, which can be attributed to water loss, and the degradation of the plant cell wall, which is observed as changes in the RSC.

Table 2. The effects of evaluation of the textural changes during the ripening time.

Time of Ripening (Days)									
0	**1**	**3**	**7**	**14**	**21**	**28**	**35**	**42**	**49**
Hardness (N)									
2.02 ± 0.23^{A}	5.74 ± 0.98^{B}	6.14 ± 0.95^{C}	15.18 ± 0.21^{D}	9.79 ± 0.58^{E}	11.09 ± 1.28^{F}	8.03 ± 1.49^{G}	10.79 ± 1.49^{H}	10.03 ± 1.19^{H}	8.99 ± 1.35^{G}
Springiness (N)									
0.95 ± 0.03^{A}	0.67 ± 0.16^{B}	0.98 ± 0.15^{A}	1.99 ± 0.36^{C}	1.06 ± 0.24^{D}	1.57 ± 0.72^{E}	2.85 ± 0.23^{F}	1.65 ± 0.81^{G}	2.11 ± 0.93^{H}	1.01 ± 0.87^{I}
Gumminess (N)									
1.03 ± 0.03^{A}	0.81 ± 0.23^{A}	1.11 ± 0.14^{A}	3.36 ± 0.28^{B}	2.46 ± 0.22^{BC}	3.12 ± 0.50^{BC}	2.18 ± 0.36^{C}	2.64 ± 0.75^{BC}	2.89 ± 1.20^{BC}	2.26 ± 1.20^{C}
Chewiness (N)									
0.98 ± 0.03^{AB}	0.53 ± 0.23^{A}	1.09 ± 0.14^{AB}	7.73 ± 1.28^{D}	2.59 ± 0.22^{ABC}	5.30 ± 0.50^{C}	6.19 ± 0.36^{C}	4.78 ± 0.75^{BC}	6.59 ± 1.20^{D}	2.41 ± 0.20^{ABC}
Cohesiveness (N)									
0.47 ± 0.03^{A}	0.13 ± 0.16^{B}	0.16 ± 0.20^{B}	0.19 ± 0.91^{C}	0.24 ± 0.60^{D}	0.26 ± 0.21^{E}	0.24 ± 0.95^{F}	0.22 ± 0.97^{F}	0.26 ± 0.66^{D}	0.22 ± 0.77^{G}

Means with different letters (A–I) in the same raw are significantly different at $p < 0.05$.

4. Conclusions

Considering the growing interest in plant-based cheese alternatives, the presented results can be useful not only from a scientific point of view, but can have wider application. Our study provides information on the microbiological, physicochemical, and textural changes in new lupine-based cheese-like products. The obtained products were stored for seven weeks and showed an abundant content of microorganisms and bioactive substances. The results presented the mechanism of cheese-like product ripening and identified critical points where textural and biochemical changes could be observed. However, future analyses using different strains and advanced proteolysis and lipolysis assays are needed for a deeper understanding of the ripening processes. In the future, plant-based cheeses may become an important part of the diet for consumers who follow a plant-based or predominantly plant-based diet, or who seek to reduce their intake of animal products. However, further in-depth studies on ripening processes with special attention to proteolysis, lipolysis, the formation of flavor and aroma compounds, consumer perception, and post-consumption effects should be conducted.

Author Contributions: Ł.Ł.: Conceptualization, investigation, methodology, formal analysis, writing—original draft preparation, supervision, project administration, funding acquisition; N.Ś., E.L. and K.M.-Z.: investigation; A.G.: formal analysis; E.D. formal analysis, visualization. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the National Science Centre (Grant no. MINIATURA 2021/05/X/NZ9/01353).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Montemurro, M.; Pontonio, E.; Coda, R.; Rizzello, C.G. Plant-Based Alternatives to Yogurt: State-of-the-Art and Perspectives of New Biotechnological Challenges. *Foods* **2021**, *10*, 316. [CrossRef] [PubMed]
2. Grossmann, L.; McClements, D.J. The science of plant-based foods: Approaches to create nutritious and sustainable plant-based cheese analogs. *Trends Food Sci. Technol.* **2021**, *118*, 207–229. [CrossRef]
3. Pua, A.; Tang, V.C.Y.; Goh, R.M.V.; Sun, J.; Lassabliere, B.; Liu, S.Q. Ingredients, Processing, and Fermentation: Addressing the Organoleptic Boundaries of Plant-Based Dairy Analogues. *Foods* **2022**, *11*, 875. [CrossRef] [PubMed]
4. Wang, J.; Li, L. Comparative Study of Chemical Composition and Texture Profile Analysis between Camembert Cheese and Chinese Sufu. *Biotechnol. Front.* **2012**, *1*, 1–8.
5. Łopusiewicz, Ł.; Drozłowska, E.; Tarnowiecka-Kuca, A.; Bartkowiak, A.; Mazurkiewicz-Zapałowicz, K.; Salachna, P. Biotransformation of Flaxseed Oil Cake into Bioactive Camembert-Analogue Using Lactic Acid Bacteria, *Penicillium camemberti* and *Geotrichum candidum*. *Microorganisms* **2020**, *8*, 1266. [CrossRef] [PubMed]
6. Fu, W.; Yano, H. Development of "New" Bread and Cheese. *Processes* **2020**, *8*, 1–23.
7. Boukind, S.; Bouaouina, J.; Bouras, H.; Benhamou, A.A.; Ablouh, E.-H.; Kassab, Z.; Khouloud, M.; El Achaby, M.; Sehaqui, H. Powerful cellulose phosphorylation by fertilizer-grade phosphate enables excellent methylene blue paper sorbent. *Int. J. Biol. Macromol.* **2022**, *219*, 949–963. [CrossRef]
8. Kamath, R.; Basak, S.; Gokhale, J. Recent trends in the development of healthy and functional cheese analogues-a review. *LWT* **2021**, *155*, 112991. [CrossRef]
9. Pipoyan, D.; Stepanyan, S.; Stepanyan, S.; Beglaryan, M.; Costantini, L.; Molinari, R.; Merendino, N. The Effect of Trans Fatty Acids on Human Health: Regulation and Consumption Patterns. *Foods* **2021**, *10*, 2452. [CrossRef]
10. Sharma, P.; Sharma, D.; Amin, A. Development of a functional fermented peanut-based cheese analog using probiotic bacteria. *BioTechnologia* **2018**, *99*, 435–441. [CrossRef]
11. Mohamed, A.; Shalaby, S.; Gafour, W.A. Quality Characteristics and Acceptability of an Analogue Processed Spreadable Cheese Made with Carrot Paste (*Daucus carota* L.). *Int. J. Dairy Sci.* **2016**, *11*, 91–99. [CrossRef]
12. Awad; Salama, W.M.; Farahat, A.M. Effect of lupine as cheese base substitution on technological and nutritional properties of processed cheese analogue. *Acta Sci. Pol. Technol. Aliment.* **2014**, *13*, 55–64. [CrossRef] [PubMed]
13. Martínez-Villaluenga, C.; Zieliński, H.; Frias, J.; Piskula, M.K.; Kozłowska, H.; Vidal-Valverde, C. Antioxidant capacity and polyphenolic content of high-protein lupin products. *Food Chem.* **2009**, *112*, 84–88. [CrossRef]
14. Abdel–Salam, A.F.; Ali, J.B.; Zayan, A.F. Effect of lupine powder on rheological, chemical and microbiological properties of yoghurt. *J. Food Dairy Sci.* **2015**, *6*, 531–546. [CrossRef]
15. Al-Saedi, N.; Agarwal, M.; Ma, W.; Islam, S.; Ren, Y. Proteomic Characterisation of Lupin (*Lupinus angustifolius*) Milk as Influenced by Extraction Techniques, Seed Coat and Cultivars. *Molecules* **2020**, *25*, 1782. [CrossRef]
16. Bartkiene, E.; Sakiene, V.; Bartkevics, V.; Juodeikiene, G.; Lele, V.; Wiacek, C.; Braun, P.G. Modulation of the nutritional value of lupine wholemeal and protein isolates using submerged and solid-state fermentation with *Pediococcus pentosaceus* strains. *Int. J. Food Sci. Technol.* **2018**, *53*, 1896–1905. [CrossRef]
17. Xia, Y.; Yuan, R.; Weng, S.; Wang, G.; Xiong, Z.; Zhang, H.; Song, X.; Liu, W.; Ai, L. Proteolysis, lipolysis, texture and sensory properties of cheese ripened by *Monascus fumeus*. *Food Res. Int.* **2020**, *137*, 109657. [CrossRef]
18. Chen, L.; Liu, H. Effect of emulsifying salts on the physicochemical properties of processed cheese made from Mozzarella. *J. Dairy Sci.* **2012**, *95*, 4823–4830. [CrossRef]
19. Zhang, X.; Esmail, G.A.; Alzeer, A.F.; Arasu, M.V.; Vijayaraghavan, P.; Choi, K.C.; Al-Dhabi, N.A. Probiotic characteristics of *Lactobacillus* strains isolated from cheese and their antibacterial properties against gastrointestinal tract pathogens. *Saudi J. Biol. Sci.* **2020**, *27*, 3505–3513. [CrossRef]
20. Leclercq-Perlat, M.-N.; Corrieu, G.; Spinnler, H.-E. Controlled production of camembert-type cheeses: Part III role of the ripening microflora on free fatty acid concentrations. *J. Dairy Res.* **2007**, *74*, 218–225. [CrossRef]
21. Mei, J.; Guo, Q.; Wu, Y.; Li, Y.; Yu, H. Study of proteolysis, lipolysis, and volatile compounds of a Camembert-type cheese manufactured using a freeze-dried Tibetan kefir co-culture during ripening. *Food Sci. Biotechnol.* **2015**, *24*, 393–402. [CrossRef]
22. Oyeyinka, A.T.; Odukoya, J.O.; Adebayo, Y.S. Nutritional composition and consumer acceptability of cheese analog from soy and cashew nut milk. *J. Food Process. Preserv.* **2019**, *43*, e14285. [CrossRef]
23. Eudier, H.; Ben-Harb, S.; Lorand, J.; Duthil, F.; Negahban, M.; Saiter, J.; Chan-Huot, M. Properties Of An Analogue Cheese Obtained From Raw Peanut. *Peanut Sci.* **2020**, *47*, 81–88. [CrossRef]
24. Mohamed, A.G.; Shalaby, S.M. Texture, Chemical Properties and Sensory Evaluation of a Spreadable Processed Cheese Analogue Made with Apricot Pulp (*Prunus armeniaca* L.). *Int. J. Dairy Sci.* **2016**, *11*, 61–68. [CrossRef]
25. Pastor-Cavada, E.; Juan, R.; Pastor, J.E.; Alaiz, M.; Vioque, J. Analytical nutritional characteristics of seed proteins in six wild *Lupinus* species from Southern Spain. *Food Chem.* **2009**, *117*, 466–469. [CrossRef]
26. Torres, A.; Frias, J.; Vidal-Valverde, C. Changes in chemical composition of lupin seeds (*Lupinus angustifolius*) after selective α-galactoside extraction. *J. Sci. Food Agric.* **2005**, *85*, 2468–2474. [CrossRef]
27. Oomah, B.D.; Tiger, N.; Olson, M.; Balasubramanian, P. Phenolics and Antioxidative Activities in Narrow-Leafed Lupins (*Lupinus angustifolius* L.). *Mater. Veg.* **2006**, *61*, 86–92. [CrossRef]

28. Wäsche, A.; Müller, K.; Knauf, U. New processing of lupin protein isolates and functional properties. *Food/Nahrung* **2001**, *45*, 393–395. [CrossRef]
29. Łopusiewicz, Ł.; Kwiatkowski, P.; Drozłowska, E. Production and Characterization of Yogurt-Like Fermented Beverage Based on Camelina (*Camelina sativa* L.) Seed Press Cake. *Appl. Sci.* **2022**, *12*, 1085. [CrossRef]
30. Łopusiewicz, Ł.; Śmietana, N.; Paradowska, D.; Drozłowska, E. Black Cumin (*Nigella sativa* L.) Seed Press Cake as a Novel Material for the Development of New Non-Dairy Beverage Fermented with Kefir Grains. *Microorganisms* **2022**, *10*, 300. [CrossRef]
31. Łopusiewicz, Ł.; Drozłowska, E.; Siedlecka, P.; Mężyńska, M.; Bartkowiak, A. Preparation and characterization of novel flaxseed oil cake yogurt-like plant milk fortified with inulin. *J. Food Nutr. Res.* **2020**, *59*, 61–70.
32. Łopusiewicz, Ł.; Drozłowska, E.; Siedlecka, P.; Mężyńska, M.; Bartkowiak, A.; Sienkiewicz, M.; Zielińska-Bliźniewska, H.; Kwiatkowski, P. Development, Characterization, and Bioactivity of Non-Dairy Kefir-Like Fermented Beverage Based on Flaxseed Oil Cake. *Foods* **2019**, *8*, 544. [CrossRef]
33. Rodriguez-Martin, N.M.; La Paz, S.M.-D.; Toscano, R.; Grao-Cruces, E.; Villanueva, A.; Pedroche, J.; Millan, F.; Millan-Linares, M.C. Hemp (*Cannabis sativa* L.) Protein Hydrolysates Promote Anti-Inflammatory Response in Primary Human Monocytes. *Biomolecules* **2020**, *10*, 803. [CrossRef] [PubMed]
34. Łopusiewicz, Ł.; Drozłowska, E.; Trocer, P.; Kwiatkowski, P.; Bartkowiak, A.; Gefrom, A.; Sienkiewicz, M. The Effect of Fermentation with Kefir Grains on the Physicochemical and Antioxidant Properties of Beverages from Blue Lupin (*Lupinus angustifolius* L.) Seeds. *Molecules* **2020**, *25*, 5791. [CrossRef] [PubMed]
35. Jiménez-Martínez, C.; Hernández-Sánchez, H.; Dávila-Ortiz, G. Production of a yogurt-like product from *Lupinus campestris* seeds. *J. Sci. Food Agric.* **2003**, *83*, 515–522. [CrossRef]
36. Zaouadi, K.N.; Ziane-Zafour, A.A.H.; Ouazib, A.M.; Arab, A.Y.; Hacini, K.; Aslan, S.S. Formulation and Optimization by Experimental Design of Dairy Dessert Based on *Lupinus albus* L. and *Stevia rebaudiana* Extracts. *Asian J. Dairy Food Res.* **2019**, *38*, 281–287. [CrossRef]
37. Guo, C.; Yang, J.; Wei, J.; Li, Y.; Xu, J.; Jiang, Y. Antioxidant activities of peel, pulp and seed fractions of common fruits as determined by FRAP assay. *Nutr. Res.* **2003**, *23*, 1719–1726. [CrossRef]
38. Lee, J.-S.; Bae, I. Quality Characteristics, Changes in Physiochemical Properties and Functional Properties of Camembert Cheese Containing Red Ginseng Powder. *Korean J. Food Sci. Anim. Resour.* **2018**, *38*, 64–77. [CrossRef]
39. Chen, J.M.; Al, K.F.; Craven, L.J.; Seney, S.; Coons, M.; McCormick, H.; Reid, G.; O'Connor, C.; Burton, J.P. Nutritional, Microbial, and Allergenic Changes during the Fermentation of Cashew 'Cheese' Product Using a Quinoa-Based Rejuvelac Starter Culture. *Nutrients* **2020**, *12*, 648. [CrossRef]
40. Bartkiene, E.; Schleining, G.; Juodeikiene, G.; Vidmantiene, D.; Krungleviciute, V.; Rekstyte, T.; Basinskiene, L.; Stankevicius, M.; Akuneca, I.; Ragazinskiene, O.; et al. The influence of lactic acid fermentation on biogenic amines and volatile compounds formation in flaxseed and the effect of flaxseed sourdough on the quality of wheat bread. *LWT-Food Sci. Technol.* **2014**, *56*, 445–450. [CrossRef]
41. Trojanowska, K.; Markiewicz, M.; Kolanowska, A.; Czaczyk, K.; Mueller, A.; Gulewicz, K. Ekstrakt Łubinowy Surowiec w Biosyntezie Paszowego Drodze Mikrobioiogicznej. *Biotechnologia* **1991**, *3*, 131–136.
42. Chen, X.; Gu, Z.; Peng, Y.; Quek, S.Y. What happens to commercial camembert cheese under packaging? Unveiling biochemical changes by untargeted and targeted metabolomic approaches. *Food Chem.* **2022**, *383*, 132437. [CrossRef] [PubMed]
43. Fritsch, C.; Vogel, R.F.; Toelstede, S. Fermentation performance of lactic acid bacteria in different lupin substrates-influence and degradation ability of antinutritives and secondary plant metabolites. *J. Appl. Microbiol.* **2015**, *119*, 1075–1088. [CrossRef] [PubMed]
44. Egounlety, M.; Aworh, O. Effect of soaking, dehulling, cooking and fermentation with Rhizopus oligosporus on the oligosaccharides, trypsin inhibitor, phytic acid and tannins of soybean (*Glycine max* Merr.), cowpea (*Vigna unguiculata* L. Walp) and groundbean (*Macrotyloma geocarpa* Harms). *J. Food Eng.* **2003**, *56*, 249–254. [CrossRef]
45. Tsafrakidou, P.; Michaelidou, A.M.; Biliaderis, C.G. Fermented Cereal-based Products: Nutritional Aspects, Possible Impact on Gut Microbiota and Health Implications. *Foods* **2020**, *9*, 734. [CrossRef]
46. Abbasi-Parizad, P.; De Nisi, P.; Sciarria, T.P.; Scarafoni, A.; Squillace, P.; Adani, F.; Scaglia, B. Polyphenol bioactivity evolution during the spontaneous fermentation of vegetal by-products. *Food Chem.* **2022**, *374*, 131791. [CrossRef]
47. Ruiz-López, M.A.; Barrientos-Ramírez, L.; García-López, P.M.; Valdés-Miramontes, E.H.; Zamora-Natera, J.F.; Rodríguez-Macias, R.; Salcedo-Pérez, E.; Bañuelos-Pineda, J.; Vargas-Radillo, J.J. Nutritional and Bioactive Compounds in Mexican Lupin Beans Species: A Mini-Review. *Nutrients* **2019**, *11*, 1785. [CrossRef]
48. Gan, R.-Y.; Shah, N.P.; Wang, M.; Lui, W.Y.; Corke, H. Fermentation alters antioxidant capacity and polyphenol distribution in selected edible legumes. *Int. J. Food Sci. Technol.* **2016**, *51*, 875–884. [CrossRef]
49. Zaworska, A.; Kasprowicz-Potocka, M.; Frankiewicz, A.; Zdunczyk, Z.; Juskiewicz, J. Effects of fermentation of narrow-leafed lupine (*L. angustifolius*) seeds on their chemical composition and physiological parameters in rats. *J. Anim. Feed Sci.* **2016**, *25*, 326–334. [CrossRef]
50. Kasprowicz-Potocka, M.; Zaworska, A.; Frankiewicz, A.; Nowak, W.; Gulewicz, P.; Zduńczyk, Z.; Juśkiewicz, J. The Nutritional Value and Physiological Properties of Diets with Raw and Candida Utilis-Fermented Lupin Seeds in Rats. *Food Technol Bio-Technol* **2015**, *53*, 286–297.

51. Xia, X.; Tobin, J.T.; Sharma, P.; Fenelon, M.; McSweeney, P.L.; Sheehan, J.J. Application of a cascade membrane filtration process to standardise serum protein depleted cheese milk for cheddar cheese manufacture. *Int. Dairy J.* **2020**, *110*, 104796. [CrossRef]
52. Ropars, J.; Didiot, E.; de la Vega, R.C.R.; Bennetot, B.; Coton, M.; Poirier, E.; Coton, E.; Snirc, A.; Le Prieur, S.; Giraud, T. Domestication of the Emblematic White Cheese-Making Fungus *Penicillium camemberti* and Its Diversification into Two Varieties. *Curr. Biol.* **2020**, *30*, 4441–4453.e4. [CrossRef]
53. Barac, M.; Vucic, T.; Zilic, S.; Pesic, M.; Sokovic, M.; Petrovic, J.; Kostic, A.; Ignjatovic, I.S.; Milincic, D. The Effect of In Vitro Digestion on Antioxidant, ACE-Inhibitory and Antimicrobial Potentials of Traditional Serbian White-Brined Cheeses. *Foods* **2019**, *8*, 94. [CrossRef] [PubMed]
54. Thambiraj, S.R.; Phillips, M.; Koyyalamudi, S.R.; Reddy, N. Antioxidant activities and characterisation of polysaccharides isolated from the seeds of *Lupinus angustifolius*. *Ind. Crop. Prod.* **2015**, *74*, 950–956. [CrossRef]
55. Sánchez-Gamboa, C.; Hicks-Pérez, L.; Gutiérrez-Méndez, N.; Heredia, N.; García, S.; Nevárez-Moorillón, G.V. Microbiological Changes during Ripening of Chihuahua Cheese Manufactured with Raw Milk and Its Seasonal Variations. *Foods* **2018**, *7*, 153. [CrossRef] [PubMed]
56. Galli, B.D.; Baptista, D.P.; Cavalheiro, F.G.; Negrão, F.; Eberlin, M.N.; Gigante, M.L. Peptide profile of Camembert-type cheese: Effect of heat treatment and adjunct culture Lactobacillus rhamnosus GG. *Food Res. Int.* **2019**, *123*, 393–402. [CrossRef] [PubMed]
57. Ong, L.; Dagastine, R.R.; Kentish, S.E.; Gras, S.L. Microstructure and Composition of Full Fat Cheddar Cheese Made with Ultrafiltered Milk Retentate. *Foods* **2013**, *2*, 310–331. [CrossRef]
58. Alu'Datt, M.H.; Rababah, T.; Alhamad, M.N.; Al-Mahasneh, M.A.; Almajwal, A.; Gammoh, S.; Ereifej, K.; Johargy, A.; Alli, I. A review of phenolic compounds in oil-bearing plants: Distribution, identification and occurrence of phenolic compounds. *Food Chem.* **2017**, *218*, 99–106. [CrossRef]