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The Influence of Rice Protein, Hemp Protein and Transglutaminase Addition on the Quality of Instant Fried Noodles

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Abstract: The goal of this study was to determine the effect of rice and hemp protein addition together with transglutaminase on the quality and fat content of fried instant noodles. The following parameters have been analyzed during this research: the amount of water, fat content, hydration time, color and texture parameters. The evaluation of sensory parameters before and after cooking of instant noodles has been performed. It can be concluded that the addition of TGase significantly decreased the fat content; the recipe with a 3% hemp protein addition with TGase in an amount of 2% was the most promising solution to obtain a low-fat product. On the other hand, the better consumer acceptance levels were observed for the rice protein addition (1%) with 2% TGase. It seems that the most promising product is the sample with a 5% addition of rice protein with 2% TGase-a reduction in the fat content by 30% (from 25% to 16%). The addition of rice protein, hemp protein and TGase reduced the fat content of the instant noodles in comparison to the control sample. The additives used had an impact on the L*, a* and b* color parameters. All instant noodles obtained during the research process were characterized by a short hydration time, which did not exceed 5 min. During the sensory evaluation, analyzed samples of the instant noodles obtained scores higher than 4.2 points on a five-point scale.

Keywords: fried instant noodles; rice protein; hemp protein; transglutaminase

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1. Introduction

Instant noodles, along with bread, are one of the most consumed grain products in the world. They can act as substitutes for rice, potatoes or groats. In many Asian countries, instant noodles are an important food ingredient in the diet, representing one of the basic food types consumed in the region. Originally, instant noodles were developed in around 5000 BC in northern China, and then spread to other countries of Southeast Asia [1,2]. In 1958, Momofuku Ando invented the world's first instant noodles in Japan. These were produced on a large scale by Nissin Foods [3–5]. Instant noodles are popular in over 80 countries around the world. According to the World Instant Noodles Association (WINA), in 2020, the demand for instant noodles was 116.560 trillion servings of instant noodles worldwide, equivalent to 319 million servings per day [6]. Modern consumers lead an intense lifestyle, and they desire a convenient food on the market that can be quickly and easily prepared for consumption. Instant noodles represent such convenience foods, as a product ready to be heated or for thermal processing [7]. The advantages of instant noodles include convenience, ease and speed of preparation, single-portion packaging, sensory values (obtained in the deep-fat frying process) expected by consumers (taste, color, texture), and an affordable price [5,8]. Another advantage of instant noodles is their absorbing the taste of sauces or other additives, and coexisting well with other ingredients [4].

Instant noodles subjected to the frying process may contain nearly 20–30% of fat used for frying [9]. After placing instant noodles in hot oil, their surface temperature rises sharply. The water on the surface of the instant noodles boils immediately, which leads to drying-heat and mass exchanges take place. This process also leads to shrinkage, followed by the development of porosity and roughness on the surface. The moisture in the gelled starch granules evaporates due to the high temperatures used during the process. Then, the spaces inside the instant noodles previously occupied by moisture are partially filled with oil [10]. Instant noodles produced with a base of semolina, compared to those made of regular wheat flour, contain higher amounts of protein: the dough swells evenly, is well bound and elastic [11]. The blistering of water evaporation from the dough is decreased, contributing to reducing the fat absorption during frying [10]. The gluten it contains is denatured during frying, and disulfide bonds are formed, which are responsible for the protein cross-linking. The resulting network combined with gelatinized starch granules creates a rigid instant noodle structure [12].

In order to reduce fat absorption in fried instant noodles, the base recipe can be enriched with modified starch or maltodextrins, classified as starch fat substitutes. They shape the characteristic structure similarly to fat, while serving as filling and moisture-retaining factors [13]. The additions of potato and corn starch are used to reduce fat absorption during frying instant noodles [14,15]. Hydrocoloids [16,17], emulsifiers [18] and antioxidants [19] are also added to improve the texture and durability of instant noodles. The addition of guar gum or other hydrocolloids makes the instant noodle structure firmer, the hydration process easier, and causes less oil absorption [14,20]. Among the cellulose derivatives, carboxymethylcellulose, microcrystalline cellulose and hydroxypropylmethylcellulose are used as substancesm contributing to the reduction in fat absorption in fried instant noodles [13]. There are also attempts to reduce the fat content or functional properties in fried instant noodles by partially replacing wheat flour with oat bran [21], B-glucan of fungal origin [22], buckwheat [23], or brown rice flour [24]. The use of the enzyme transglutaminase (TGase), thanks to the cross-linking properties of most plant proteins or animal origin, affects the gluten contained in instant noodles [25]. TGase induces the formation of high-molecular polymers, thus strengthening the gluten network in instant noodles. The enzyme, when using high-quality flour containing large amounts of high-molecular glutenins, increases protein polymerization. Improving the protein network in instant noodles extends its freshness, simultaneously maintaining the original sensory characteristics. Another important feature of TGase affecting the quality of instant noodles is the higher degree of water binding [26]. TGase is used for the cross-linking of milk proteins, soy proteins, meat proteins, and the production of bread or yoghurt. This enzyme may also have beneficial effects on the texture-improving the hardness and the chewiness [25,27], and the volume and the internal structure of bread, cookies, cakes and instant noodles [28–31]. High fat contents in instant noodles adversely affect the health value and limit the shelf life of the product due to the changes in fats during storage [10].

Attempts to increase the health value of instant noodles involve the addition of components such as various types of proteins (e.g., hemp and rice proteins) serving as factors to decrease the fat content.

Hemp protein is an excellent source of nutrients in the human diet; this protein is rich in exogenous amino acids [32,33]. Wang et al. (2008) conducted a study of the amino acid composition of a hemp protein isolate. Based on their results, it was found that it is rich in arginine, aspartic acid, glutamic acid, serine, and leucine. Hemp seed protein is also a source of bioactive peptides. These peptides are active in vitro and in vivo as antioxidant, antihypertensive, anticoagulant, antibacterial and hypocholesterolemic factors. They are also characterized by the ability to chelate metal ions, remove toxic free radicals and inhibit the oxidation of linoleic acid [34].

A rice protein is a vegetarian and gluten-free protein, with low fat and cholesterol contents [35]. The protein is characterized by a well-balanced amino acid composition, as well as representing a good source of exogenous amino acids [36]. Rice protein has a high content of glutamic acid, aspartic acid, arginine and leucine, and contains methionine, cysteine and small amounts of lysine [35]. It is characterized by good health properties, exhibits hypoallergenic, hypolipidemic, hypocholesterolemic and anti-cancer effects, and prevents obesity [37].

The factors mentioned above have led to research to reduce fat absorption during frying, and justify deep studies to enrich the basic recipe composition of instant noodles with the addition of hemp and rice proteins in combination with TGase. Bearing in mind the ability to improve the health value of the additions described above, the aim of this study was to determine the effect of rice and hemp protein additions together with transglutaminase on the quality characteristics of fried instant noodles, while decreasing the fat content.

2. Materials and Methods

Semolina, coarse flour milled from tetraploid durum wheat (Triricum durum), was purchased from Bogutyn Mill (Radzyń Podlaski, Poland). The flour composition was 13% moisture, 0.62% ash, and 11% protein. The granulation of the flour was 100% finer than 149 µm (U.S. 100 mesh).

The food-grade samples of TGase were provided by Barentz LLC/Roquette (Warsaw, Poland). The samples of rice protein were provided by Bene Vobis, (Gdansk, Poland), and hemp protein was provided by Intenson (Karczew, Polska). All samples were in powder state, stored in a cool and dry warehouse until use. As a frying medium, refined rapeseed oil was used, (ZT Kruszwica S.A., Kruszwica, Poland), with a smoke point of 204 °C (399.2 F).

2.1. Preparation of Instant Fried Noodles

The instant noodle dough was formulated by mixing 100% flour, 3% (kg/kg flour) salt, 1–5% (kg/kg flour) rice protein, 1–5% (kg/kg flour) hemp protein, 1–2% (kg/kg flour) TGase, and 35% (kg/kg flour) water. The water solution of dissolved salt, rice and hemp protein was added to the flour (150 g) and formed into a dough. The crumbly dough was then placed into resealable plastic bags and rested for 30 min before further size reduction.

The dough was then passed through the roller unit attachment of an instant noodles machine (Kitchen Aid, Benton Harbor, MI, USA) with the regulating knob set at No.1 position (2.5 mm). The resulting sheet was folded in half and passed again through the rollers. This process was repeated several times until the dough sheet was smoothly formed. The thickness of the sheet was reduced stepwise by passing between the rollers of the instant noodle machine. The final cutting roll gap was adjusted to 1.0 mm and the noodle sheet was cut through a cutter attachment. The instant noodles were then steamed in a steamer for 5 min. The final step was frying in rapeseed oil (ZT Kruszwica S.A., Kruszwica, Poland) at a temperature of 170 °C for 90 s. After the frying process, the noodle strands were cooled at room temperature (~23 °C) and packed into polyethylene bags.

The instant noodles were made on the basis of semolina flour from the Triticum Durum wheat variety (Bogutyn Młyn, Radzyń Podlaski, Poland) according to the recipe developed by Kubomura [9], including: 150 g of semolina flour, 35% water in relation to the amount of semolina and 3 g of cooking salt (Cenos Sp. Zoo, Września, Poland). The commercial samples of hemp protein (Intenson, Karczew, Poland), rice protein (Bene Vobis, Gdańsk, Poland), and the enzyme TGase (EC 2.3.2.13) by Brenntag Polska (Kędzierzyn-Koźle, Poland) were used as functional additives, influencing the physicochemical properties of instant noodles.

The following variants were developed in the study: a control sample, a test sample with the use of hemp protein in amounts of 1%, 2%, 3%, 4% and 5%, rice protein in amounts of 1%, 2%, 3%, 4% and 5% and the enzyme TGase in amounts of 1% and 2% (in relation to the flour weight).

The control sample was prepared without additives. Other variants were prepared as follows: 5 variants of instant noodles were prepared with the addition of hemp protein, 5 variants with the addition of rice protein, 10 variants with the addition of hemp protein and TGase, and 10 variants with the addition of rice protein and TGase. The production of instant noodles began with weighing out the basic recipe ingredients. Salt was dissolved in water at a temperature of about 25–30 °C. The semolina was mixed with water and the dough was kneaded by hand for 20 min. The dough obtained in this way was rolled out, each time reducing the distance between the rolls. A Kitchen AID (K5SS Heavy Duty, Benton Harbor, USA) robot was used for rolling and cutting the dough. In the next step, the dough sheets were cut into 1.5 mm wide instant noodles threads. Instant noodle evaporation was carried out for 5 min; then, the instant noodles were fried in a Moulinex Uno (Mayenne, France) fryer, in rapeseed oil (ZT "Kruszwica" S.A., Kruszwica, Poland), for 90 s at a temperature of 170 °C. The noodles were then drained, cooled and packed in polyethylene bags.

2.2. Moisture Analysis

Moisture contents were measured by slightly modifying the air oven method [38], and each sample was prepared in triplicates. Approximately 5 g instant noodle pieces were accurately weighted into the pre-weighted dishes and placed into the oven with the lids placed under the respective dishes. These samples were dried at 100 ± 1 °C for 2 h and cooled in a desiccator for 30 min. The process of drying, cooling and weighting was repeated until constant weight obtained. The results were calculated in percentages using the following equation:

$$\text{Moisture content (\%)} = (\text{Loss in weight of dish, lid and sample}/\text{Initial weight of sample}) \times 100$$

2.3. Fat Analysis

Fat contents of the instant noodles were determined by automated Soxhlet extraction (Soxtec™ 2050 Auto Fat Extraction System, Hilleroed, Denmark). The samples of instant noodles (3 g) were weighed into thimbles and inserted into the extraction unit. Solvent (petroleum ether 40/60 pa) was added to the extraction cups in a closed system. Three repetitions were performed for each measurement and the average data collected were used for statistical analysis. The dried samples were accurately re-weighted, and the fat contents were calculated using the following equation:

$$\text{Fat content (\%)} = (\text{Initial weight of sample} - \text{final weight of sample}/\text{Initial weight of sample}) \times 100$$

2.4. Color Determination (L, a*, b* Parameters)*

Characterization of the instant noodles' color was performed using the L*a*b* system proposed by the International Commission on Illumination (CIE) in the work of Papadakis, Abdul-Malek, Kamdem, and Yam (2000). L* refers to the luminosity or lightness component, and a* (intensity of red (+) and green (−)) and b* (intensity of yellow (+) and blue (−)) are the chromaticity coordinates. All sampled instant noodles were analyzed in terms of the referred parameters using a Minolta CR-310 colorimeter (Konica-Minolta, Osaka, Japan), which was calibrated prior with a white standard tile. Five repetitions were performed for each measurement.

2.5. Texture

The measurements of hardness and adhesiveness were carried out 10 min after the hydration of instant noodles at a temperature of 20 ± 2 °C. The samples were analyzed on a TA.XT Plus Analyzer (Stable Micro Systems, Godalming, UK). The hardness and the adhesiveness were determined using a P/36R probe with a diameter of 36 mm. The compression speed during the test was 2 mm·s^{-1}, until 75% deformation was obtained. The determinations were performed in five replications.

Firmness was measured with a TA-XT Plus Analyzer (Stablo Micro Systems, Godalming, UK) using an A/LKB-F head. The travel speed of the measuring element used in the test of instant noodles was 0.17 mm·s^{-1}, with a distance of 4.5 mm. The determination was performed in five replications.

2.6. Hydration Time

In order to determine the hydration time of instant noodles, boiling water was poured over the sample and the noodle hardness was checked using sensory means at intervals of 1 min. The instant noodle hardness was determined using the following markings: "−"-hard instant noodle, "−/+"-semi-soft instant noodle, "+"-soft instant noodle, "++"-very soft instant noodle [39].

2.7. Sensory Evaluation

Sensory evaluations of the 31 instant noodle samples were performed in accordance with the Polish Standard PN-A-74131: 1999 "Pasta", on a five-point scale. The sensory features were analyzed by a trained team of 10 persons. The analysis was performed in two stages: before and after hydration. In the first stage, the appearance and smell of the instant noodle were assessed before cooking. Before the second stage, hot water was poured onto the instant noodles, which were then left in boiling water for 3 min. The noodles were taken out and left for 10 min again. In the second step, the appearance, smell and taste were analyzed. The quality characteristics of the fried instant noodles were assessed on a scale of 1 to 5. Results for each feature were presented as the mean of grade point.

2.8. Statistical Analysis

Data from the physico-chemical and instrumental analysis were subjected to two-way ANOVA. Bonferroni corrections were applied to all ANOVA results. Sensory test result analysis was performed using Mann–Whitney U tests. The results of the QDP were assessed based on principal component analysis (PCA). The statistical significance was recognized when $p < 0.05$. All tests were performed using STATISTICA 13.3 PL software (StatSoft, Kraków, Poland).

3. Results and Discussion

Water migrates from the surface of instant noodles towards the center, affecting the structure after pouring boiling water over the instant noodle. According to Ding and Yang [18], the shorter the hydration time, the better the quality of instant noodles. The authors suggested that the rehydration time in 80 °C water should be less than 360 s; otherwise, instant noodles are not considered instantaneous. The instant noodles obtained in the work had a hydration time within this range (Tables 1 and 2). However, the higher concentrations of additives used increased the hydration time of the instant noodles [26].

3.1. Water Content

Water content is a very important parameter related to food quality, and in the case of instant noodles, it affects its storage suitability [40]. During the frying process of instant noodles, the water content decreased from an initial 30–40% to a final 2–5% [3,20]. The obtained results of the water content of instant noodles with the additions of rice protein, rice protein and TGase were in the range of 2.33–4.29%, and in the case of instant noodles with the addition of hemp protein, hemp protein and TGase was in the range from 2.33% to 4.36%. The above results follow acceptable standards: according to Kim et al. [41], the water content of instant noodles should be within the range from 3% to 5%.

Mohammadi et al. [42] obtained similar relationships, and according to the authors, the cross-linking of glutamine and lysine with TGase may lead to water binding and increases the ability to retain moisture in the dough.

A significant effect of the addition of rice protein, hemp protein and TGase on the values of water content was found (Figure 1). The water content of the control sample was 2.33%. It was noticed that regardless of the type and amount of additive used, both instant noodles with the addition of rice protein, as well as instant noodles with the addition of hemp protein, contained more water than the control sample.

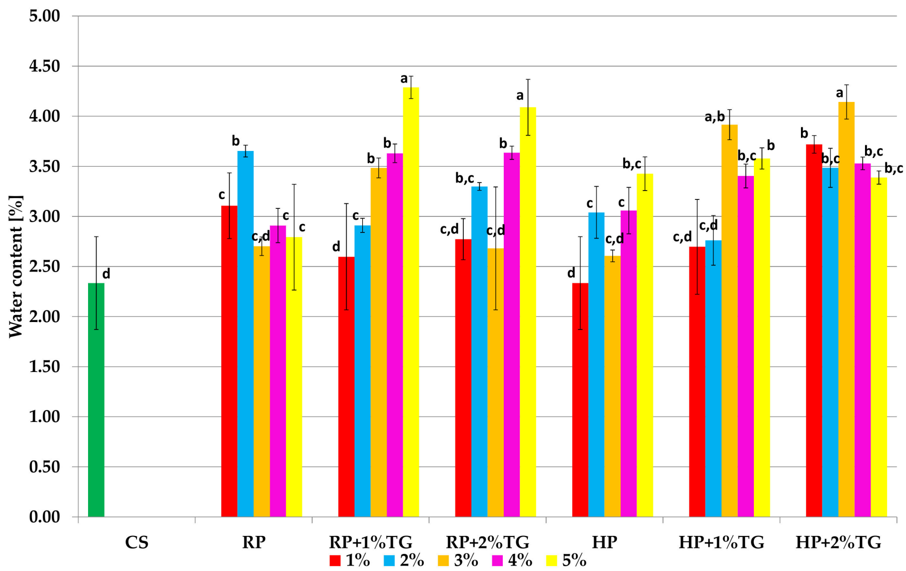

Figure 1. Average values of water contents in instant noodles with the additions of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**d**) represent statistically significant differences between variables (water content and samples) ($p < 0.05$).

The water content in instant noodles with the addition of rice protein, rice protein and TGase ranged from 2.60% to 4.29%. The highest water content was found in instant noodles with a 5% addition of rice protein and 1% addition of TGase. In the case of these instant noodles, it was observed that with the increase in the protein addition and the 1% addition of TGase, the water content increased. The addition of TGase increased the water content in instant noodles with 4% and 5% protein.

Instant noodles with the addition of hemp protein and hemp protein with TGase contained 2.61% to 4.36% water. The highest water content was obtained for instant noodles with 5% hemp protein and 2% TGase. It was observed that the addition of TGase increased the water content in instant noodles with the addition of hemp protein.

The obtained water content results were similar to the results obtained by Rekas and Marciniak-Lukasiak [16], which ranged from 2.67% to 4.10%. According to Sobota and Luczak [43], the water content in instant noodles should be from 2.73% up to 9.44%.

According to Gerrard et al. [44], elimination of the amide group from glutamine (resulting from the activity of TGase) and its conversion to glutamic acid may result in increased water absorption. Glutamic acid reduces hydrophobicity, resulting in increased water absorption. On the other hand, Mohammadi et al. [42] believe that the cross-linking of glutamine and lysine by TGase may lead to water binding and increase the moisture retention capacity in the dough.

According to the standard [45], the water content in instant noodles should be below 12.5%. A higher value may cause mold and fungus growth [40]. On the basis of the conducted research, it was found that all instant noodles had a water content consistent with the requirements stated in the standard.

3.2. Fat Content

The fat content is an important distinguishing feature of the quality of fried instant noodles. The fat used for frying is a heat carrier; however, due to migration into the internal structure of instant noodles, becomes a new ingredient [10]. The final fat content in the finished product depends on the quality of the raw materials used in the production of instant noodles and the parameters of the frying process (temperature, time) [46]. High fat contents increase the production costs of the product, and also shorten the use-by date [15].

Differences in the fat content in the process of frying instant noodles result from the microporous structure of the instant noodles and the amount of water absorbed in the evaporation process [10]. In the frying process, water contained in the food migrates from the center of the product to its surface. As a result of the evaporation of water from the inside of the instant noodle threads, a porous structure is formed. Water evaporating leaves empty spaces in the microporous structure of instant noodles, which fill with fat during frying [10]. The use of protein preparations has a beneficial effect due to the reduced fat content and the possible increase in nutritional value resulting from the properties of the added raw materials. According to Kuraishi et al. [47], the addition of TGase to the dough can reduce fat absorption by up to 25%.

On Figure 2, the obtained results of the fat content measurements are presented. It was found that the addition of hemp protein, rice protein and TGase significantly influenced the value of this parameter. The fat content in the control sample without additives was 25.59%. Regardless of the type and amount of the addition, both instant noodles with the addition of rice protein and noodles with the addition of hemp protein contained less fat compared to the control sample. According to Park and Baik [48], the protein content influences the fat absorption during instant noodle frying. A high protein content results in reduced fat absorption.

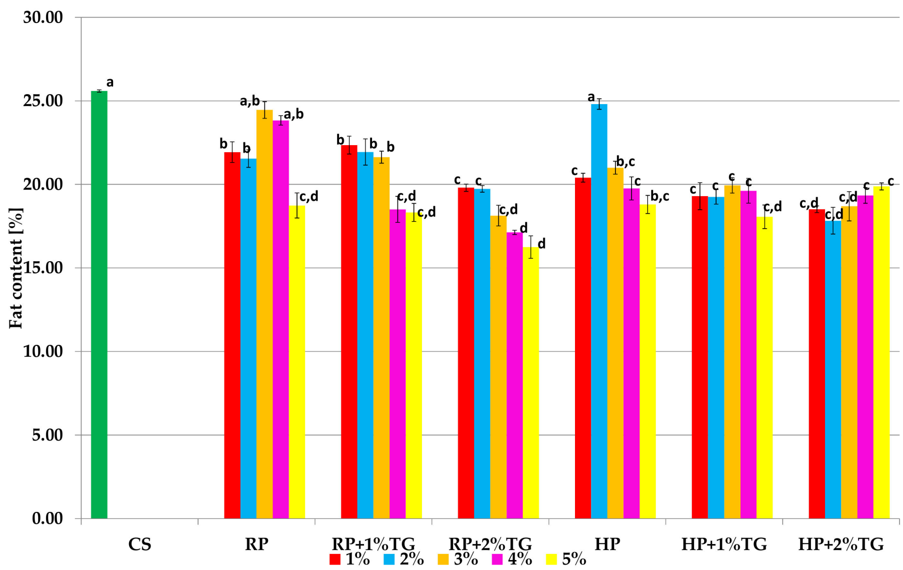

Figure 2. Average values of fat content in instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various addition of rice protein. Mean values denoted by different letters (**a**–**d**) represent statistically significant differences between variables ($p < 0.05$).

The instant noodles with the addition of rice protein and rice protein with TGase contained 16.25% to 24.46% fat. The lowest fat content was found in instant noodles with a 5% addition of rice protein and 2% addition of TGase. In the case of instant noodles with 1% and 2% additions of TGase, it was noticed that with the increase in the amount of additive used, the fat content decreased.

The fat contents in instant noodles with the additions of hemp protein and hemp protein and TGase ranged from 17.83% to 24.81%. The lowest fat content was found in instant noodles with 2% additions of both hemp protein and TGase. A high amount of fat, comparable to the control sample (25.59%), was obtained in the case of instant noodles with 2% protein.

One can notice that in most cases, the addition of TGase had an effect on reducing the fat content in instant noodles with the addition of rice protein and hemp protein. This may be observed because the action of TGase has resulted in a stronger and less porous protein network in a cake and contributes to lower oil absorption by the product during frying [12].

According to Galiński et al. [49], the fat content should be within the range 20–30%. However, Hatcher [2] claimed that instant noodles can contain 15–40% fat. All analyzed instant noodles were characterized by fat contents in this range.

Kuraishi et al. [47] investigated the effect of the enzyme preparation on the amount of fat absorbed during frying. They suggest that addition of TGase in dough reduces fat absorption by 25%. In the conducted research, the addition of TGase reduced fat absorption by 13–36.5% in instant noodles with the addition of rice protein, and by 22–30% in samples with the addition of hemp protein.

In the case of fried instant noodles, there is a correlation between the water content and the fat content. As the water content increases, the fat content decreases. According to the mass balance, the amount of fat absorbed is equal to the amount of water evaporated. During the frying process, the water contained in the product moves from the center to its surface. Evaporation of the water from the inside leads to the formation of a porous structure. The water evaporates and leaves spaces in the microporous structure, which are filled with fat during frying [10].

3.3. Color

Color is an important determinant of food quality because it affects consumer acceptability [50]. The color may be influenced by the conditions of the technological process, mainly the frying parameters, e.g., temperature, time, and type of frying medium, as well as the recipe composition of the product [51]. In combination with texture, it also affects the appearance of instant noodles and mainly depends on the quality of the flour [52] and process parameters [39]. The protein content is also important, contributing to the characteristic yellow color of instant noodles [48]. According to Hatcher et al. [2], noodles should be light, free from discoloration and have a typical light-yellow color. Park and Baik [48] claim that the color parameters of instant noodles depend, inter alia, on the type of raw materials used. L* is a critical parameter in the frying industry, which is usually the first quality attribute judged by consumers when determining product acceptance. Low L* values represent a dark color and are mainly associated with non-enzymatic browning reactions [53]. Instant noodles made from durum wheat flour are generally brighter (L* > 76.4), whereas those made from common wheat flour are less bright (L* < 76.4). However, according to Chon-Sik Kang et al. [54], the brightness of instant noodles should be in the range 73.5–82.0.

The brightness level (parameter L*) of the obtained instant noodles ranged from 60.72 to 68.33 (Figure 3). All tested instant noodles obtained lower values than the control sample (68.33). The lowest value of the L* parameter was measured in the case of instant noodles with 5% rice protein addition. As the amount of protein addition increased, a decrease in the L* brightness level was observed.

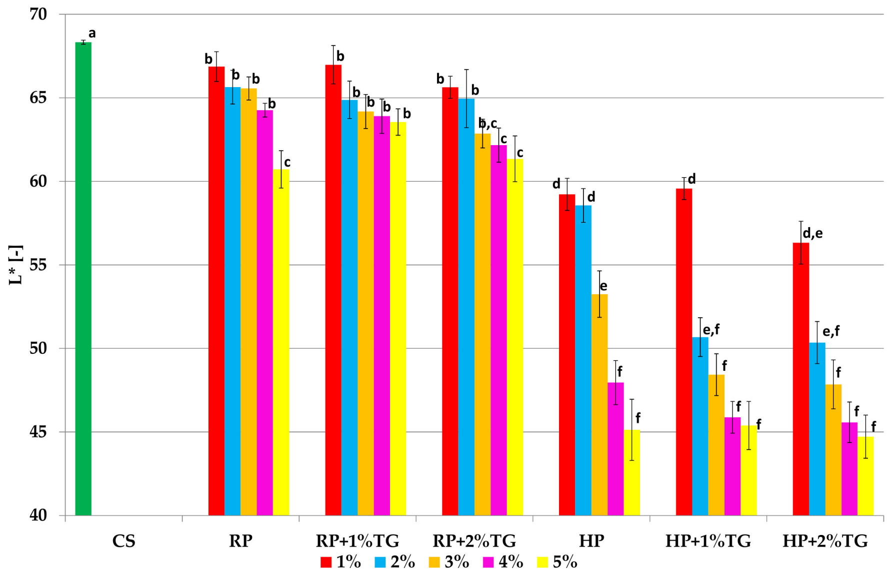

Figure 3. Average values of the L * parameter of instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**f**) represent statistically significant differences between variables ($p < 0.05$).

According to Park and Baik [48], instant noodles should have a brightness level ranging from 73.4 to 81.1. On the basis of the obtained results, it was found that they did not fall within this range. The differences may have arisen from the use of other ingredients in the instant noodle recipe.

The addition of hemp protein and hemp protein with TGase reduced the L* parameter compared to the control sample (68.33). The brightness level of instant noodles with additives ranged from 44.72 to 59.57. The lowest value of this color parameter was observed in instant noodles with 5% hemp protein addition with 2% TGase, and the highest value in instant noodles was with 1% hemp protein and 1% TGase. It was noticed that with the increase in the hemp protein addition, the value of the L* parameter decreased.

Norajit et al. [55] added hemp flour to the energy bars and Radočaj et al. [56] added hemp flour to gluten-free crackers: both noticed a decrease in the value of the L* parameter. According to Czerwińska [57], the type of flour and additives used have the greatest impact on the color of instant noodles. The darker color of the obtained instant noodles resulted from the color of the raw material used.

The value of the a* parameter ranged from −1.38 to 0.65 (Figure 4). The lowest value of this parameter was recorded in the control sample, and the highest was in the case of instant noodles with a 5% addition of rice protein. On the basis of the obtained results, it was noticed that the addition of rice protein both individually and in combination with TGase increased the value of the a* parameter compared to the control sample. The control sample, as well as instant noodles with 1–3% additions of rice protein, instant noodles with 1–3% additions of rice protein with a 1% addition of TGase, and instant noodles with 1–2% additions of rice protein and a 2% addition of TGase were characterized by a saturation of green color (negative values). Red color (positive values) was found in the remaining instant noodles.

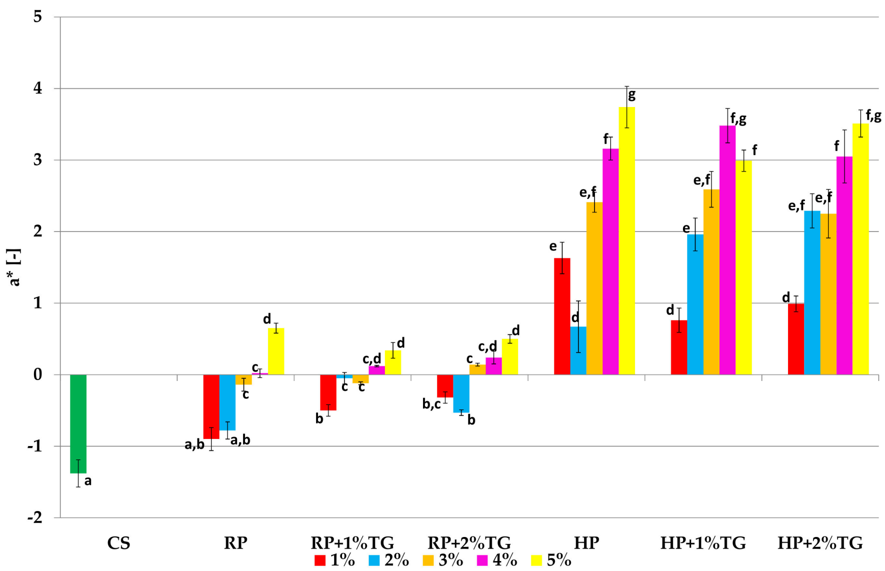

Figure 4. Average values of the a* parameter of instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**g**) represent statistically significant differences between variables ($p < 0.05$).

The addition of hemp protein and hemp protein and TGase had an impact on the value of the a* parameter of instant noodles. The lowest value of this parameter was achieved by the control sample (−1.38). For the other samples with additives, positive values of a* from 0.67 to 3.74 were observed. The applied additives caused the saturation to change from green for the control sample to red for all samples with additives. Similar observations i were noted by Perczynska and Marciniak-Lukasiak n their research [58]. The b* parameter values of the tested instant noodles with the addition of rice protein ranged from 21.87-noodles with 5% rice protein-to 23.68-control sample (Figure 5). Analysis of the value of this color parameter showed that all tested instant noodles were saturated with yellow (positive values).

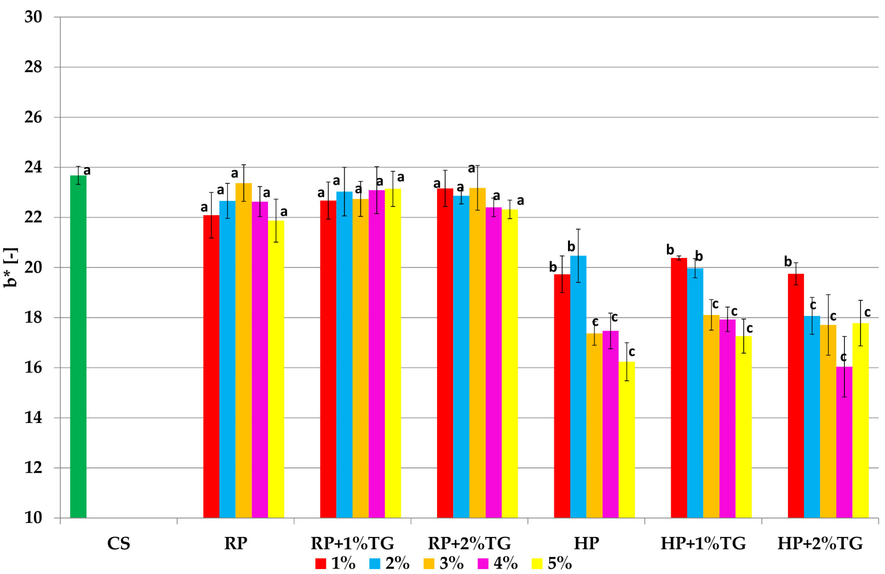

Figure 5. Average values of the b* parameter of instant noodles with the addition of rice protein,

hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**c**) represent statistically significant differences between variables ($p < 0.05$).

Based on the results of the b* parameter, it was found that the addition of hemp protein and TGase influenced the value of this color parameter. All instant noodles with additives had a lower value than the control (23.68). The instant noodles obtained positive values from 16.04 (sample with 4% hemp protein addition with 2% TGase) to 20.47 (sample with 2% hemp protein addition). These were saturated with yellow.

Marciniak-Lukasiak and Ciszek [39] and Marciniak-Lukasiak and Zbikowska [59] also obtained instant noodles with a yellow saturation.

3.4. Hydration Time

The hydration time is an important quality criterion for instant noodles. After pouring boiling water over instant noodles, the water migrates from the surface of the instant noodle towards the center, affecting its structure. The product should be ready for consumption after hydration in hot water in a few minutes. Ding and Yang [18] believe that the shorter the hydration time of an instant noodle, the better its quality. According to these authors, the hydration time of instant noodles in water at 80 °C must be less than 360 s, otherwise it is not considered instantaneous.

Table 1. Hydration times of instant noodles with the addition of rice protein, rice protein and TGase (the meaning of symbols used for evaluation is as follows: "−"-hard instant noodle, "−/+"-semi-soft instant noodle, "+"-soft instant noodle, "++"-very soft instant noodle).

	Protein Content [%]	Time [min]				
		1	2	3	4	5
Control sample	-	−	−/+	+	+	++
Rice protein	1	−	−/+	+	+	++
	2	−	−/+	+	+	++
	3	−	−/+	+	+	++
	4	−	−/+	+	+	++
	5	−	−/+	+	+	++
Rice protein and 1% TGase	1	−	−/+	+	+	++
	2	−	−/+	+	+	++
	3	−	+	+	+	++
	4	−	+	+	+	++
	5	−	−/+	+	+	++
Rice protein and 2% TGase	1	−	−/+	+	+	++
	2	−	−/+	+	+	++
	3	−	+	+	+	++
	4	−	−/+	+	+	++
	5	−	−/+	+	+	+

The hydration time of instant noodles with the addition of rice protein, rice protein and TGase was, in most cases, equal to 3 min (Table 1). Only the additions of 3% and 4% protein with 1% TGase and 3% protein addition with 2% TGase reduced the hydration time to 2 min. The instant noodles with 5% rice protein and 2% TGase were al dente after 5 min rehydration in boiling water.

The hydration times of instant noodles with the addition of hemp protein and hemp protein and TGase were 2 to 4 min (Table 2). Samples with a 1% addition of protein, 4% addition of protein and 1% addition of TGase had a 4 min hydration time. The addition of hemp protein with a 1% addition of TGase, 3% addition of protein with 2% addition of TGase reduced the hydration time to 2 min.

Similar results were obtained by Marciniak-Lukasiak and Ciszek [39] in their study over the quality of instant noodles fried in rapeseed oil. Instant noodles had a hydration time of 3 min. According to Perczyńska and Marciniak-Lukasiak [58], instant noodles can be classified as an instant product if their hydration time does not exceed 6 min. Hydration times of the tested samples with the addition of rice protein and hemp protein with TGase were within the acceptable standard and can be considered as instant noodles.

Table 2. Hydration times of instant noodles with the addition of hemp protein, hemp protein and TGase (the meaning of symbols used for evaluation is as follows: "−"-hard instant noodle, "−/+"-semi-soft instant noodle, "+"-soft instant noodle, "++"-very soft instant noodle).

	Protein Content [%]	Time [min]				
		1	2	3	4	5
Control sample	-	−	−/+	+	+	++
Hemp protein	1	−	−/+	−/+	+	++
	2	−	−/+	+	+	++
	3	−	−/+	+	+	++
	4	−	−/+	+	+	++
	5	−	−/+	+	+	++
Hemp protein and 1% TGase	1	−	+	+	+	++
	2	−	+	+	+	++
	3	−/+	+	+	+	++
	4	−	−/+	−/+	+	++
	5	−	+	+	+	++
Hemp protein and 2% TGase	1	−	−/+	+	+	++
	2	−	−/+	+	+	++
	3	−	+	+	+	++
	4	−	−/+	+	+	++
	5	−	−/+	+	+	++

3.5. Texture

The texture of instant noodles can be characterized as rubbery, firm and smooth. The texture includes parameters such as smoothness, softness, hardness, firmness, viscosity, cohesiveness, elasticity, chewiness and gumminess [3]. Instant noodles should be firm, and in some cases hard [58].

Hardness in the physical sense is the force necessary to achieve a given deformation. On the other hand, in a sensory sense, it is the force needed to compress the noodles between the molars [60,61].

Analyzing the hardness values, a significant effect of the addition of rice protein and TGase on the texture of instant noodles was found (Figure 6). The hardness ranged from 0.34 (instant noodles with a 1% addition of rice protein and a 2% addition of TGase) to 1.00 N (instant noodles with a 3% addition of rice protein). The value of this texture parameter for the control was 0.61 N. Samples with the addition of rice protein achieved hardness values higher than the control sample.

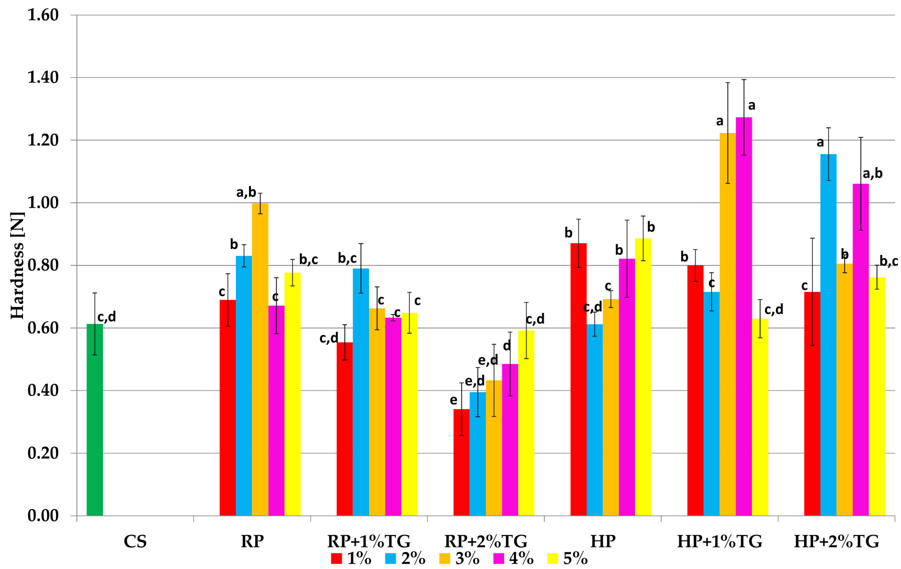

Figure 6. Average hardness values of hydrated instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**e**) represents statistically significant differences between variables ($p < 0.05$).

Compared to the control sample, a decrease in the hardness value was observed in the case of instant noodles with rice protein and 2% TGase. In the case of instant noodles with 2% TGase, the higher the level of protein added, the higher the hardness. The addition of TGase in amounts of 1% and 2% reduced the hardness of instant noodles with the addition of rice protein.

The hardness of the tested instant noodles ranged from 0.61 to 1.27 N. On the basis of the obtained results, it was found that the addition of hemp protein and TGase had a significant differentiation effect on the hardness of instant noodles. Values comparable to the hardness of the control sample (0.61 N) were obtained for samples with additions of 2% hemp protein (0.61 N) and 5% hemp protein with 1% TGase (0.63 N). It was noticed that in the cases with 2%, 3% and 4% protein additions, as well as the addition of TGase, the hardness of the instant noodles was increased. Wu and Corke [62] found a similar relationship. Studies conducted by Cierach and Grala [63], Piotrowska and Dolata [64] and Pyrcz et al. [65] showed that the addition of TGase increased the hardness of meat products.

In studies on the effect of TGase on the quality of fried instant noodles, Choy et al. [12] obtained hardness values in the range of 19.81 to 38.78 N. On the basis of the obtained results, it was found that they were much lower. Perczynska and Marciniak-Lukasiak [58] obtained instant noodles with hardness values from 2.41 to 4.28 N, and Rekas and Marciniak-Lukasiak [16] obtained instant noodles with hardness values from 4.37 to 8.16 N. The hardness of samples with the additions of rice and hemp protein did not fit in this range. The differences may have resulted from the different raw material compositions of instant noodles.

Adhesiveness in the sensory sense is the force needed to prevent instant noodles from sticking to the palate while eating. On the other hand, in the physical sense, it is the work necessary to overcome the forces between the surface of the food and the surface with which the food is in contact [60,61].

The additives used significantly influenced the adhesiveness of instant noodles, ranging from −0.85 to −0.04 g * s (Figure 7). It was observed that the additions of rice protein and TGase had differentiating effects on the adhesiveness of the instant noodle. The lowest value of adhesiveness was characteristic for instant noodles with a 4% addition of protein and 1% addition of TGase. On the other hand, the highest adhesiveness was recorded in the case of instant noodles with a 3% addition of rice protein and a 1% addition of TGase. Adhesiveness values comparable with that of the control sample (−0.70 g * s) were obtained for instant noodles with a 1% addition of rice protein (−0.72 g * sec).

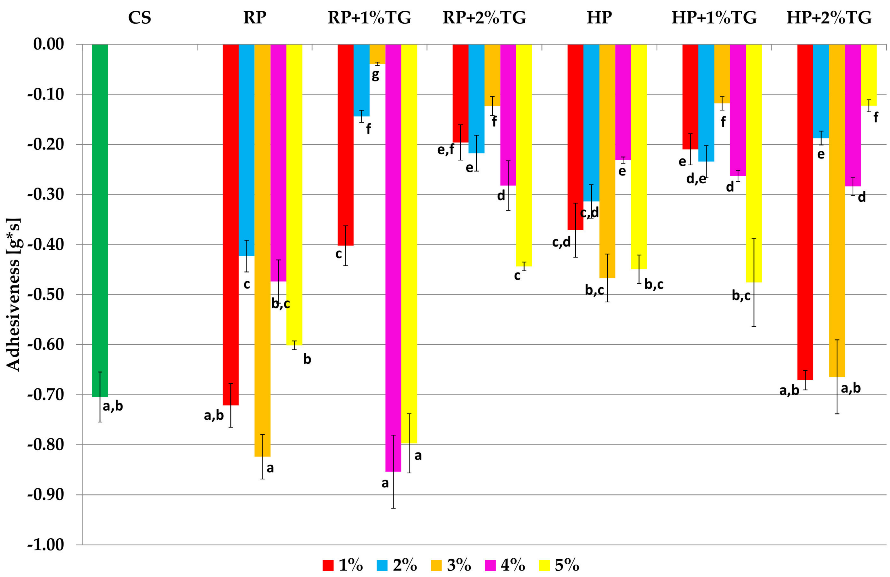

Figure 7. Average adhesiveness values of hydrated instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**g**) represent statistically significant differences between variables ($p < 0.05$).

The adhesiveness of instant noodles ranged from −0.70 to −0.12 g * s. A significant effect of the addition of hemp protein and TGase on the increase in the value of this texture parameter was noticed compared to the control sample (−0.70 g * s). The addition of protein and TGase had a differentiating effect on adhesiveness.

According to Kang et al. [66], instant noodle viscosity is defined as adhesiveness, and high values are undesirable. Choy et al. [12] conducted a study to determine the effect of TGase on the quality of fried instant noodles. They achieved adhesiveness values ranging from −0.59 to −0.14. The measurement results of the tested instant noodles are similar.

The firmness is the degree of resistance exhibited by the instant noodles during the first bite, and in a sensory sense, it is the force required to chew the instant noodles [67].

A significant effect of the addition of rice protein and TGase was observed in the case of the firmness of instant noodles (Figure 8). The firmness ranged from 0.58 to 2.81 N, and the value of this texture parameter for the control sample was 1.13 N. The lowest value was noted for instant noodles with a 5% addition of rice protein, and the highest value for instant noodles was observed with a 1% addition of rice protein with 2% TGase. In comparison to the control sample, an increase in the firmness value was found after an addition of 2% TGase to samples with the addition of rice protein.

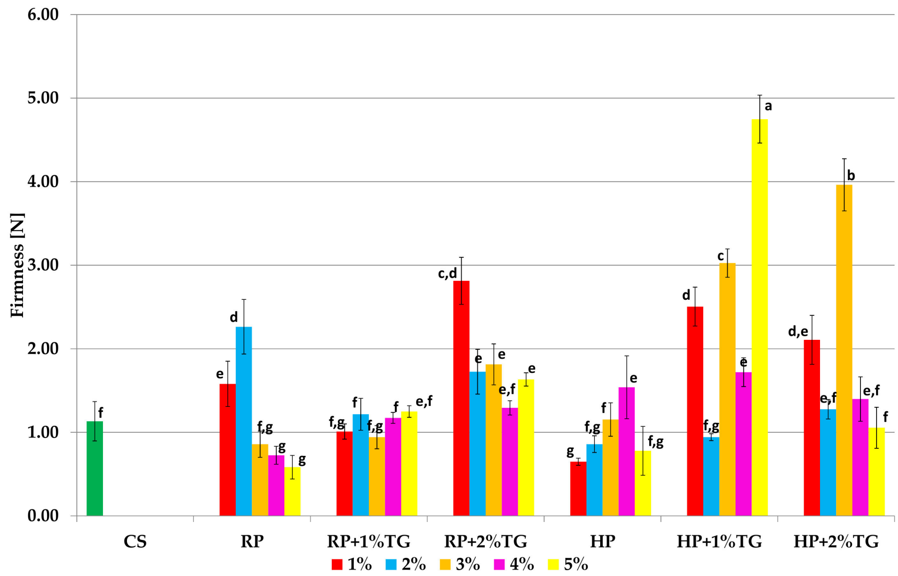

Figure 8. Average firmness values of hydrated instant noodles with the addition of rice protein, hemp protein and TGase (CS—control sample, RP—rice protein, RP+1%TG—rice protein with 1% TGase, RP+2%TG—rice protein with 2% TGase, HP—hemp protein, HP+1%TG—hemp protein with 1% TGase, HP+2%TG—hemp protein with 2% TGase). Columns correspond to various additions of rice protein. Mean values denoted by different letters (**a**–**g**) represent statistically significant differences between variables ($p < 0.05$).

Based on the analysis of the obtained results, it was found that the additives used in the study had a significant effect on the firmness values of instant noodles, ranging from 0.65 to 4.75 N. The lowest firmness was noticed in the case of instant noodles with a 1% addition of hemp protein, and the highest was in the case of instant noodles with a 5% addition of hemp protein and a 1% addition of TGase. Values comparable to the firmness value of the control sample (1.13 N) were obtained for instant noodles with a 5% addition of protein and a 2% addition of TGase (1.05 N), and instant noodles with a 3% addition of protein (1.15 N). In most cases, the addition of TGase to hemp protein caused an increase in the firmness value of instant noodles.

According to Gulia et al. [3], the addition of an enzyme preparation (e.g., TGase) increases the firmness of instant noodles. This statement was confirmed in the conducted research. Instant noodles with the addition of TGase are characterized by higher breaking strength and hardness. After cooking, instant noodles retain their firmness and elasticity [47].

3.6. Sensory Analysis

Table 3 presents the results of sensory evaluations of fried instant noodles with the addition of rice protein and rice protein with TGase.

After the sensory scoring, no significant differences were found in the results of the quality distinguishing features. The highest score was given to instant noodles with 1% and 3% additions of protein and a 2% addition of TGase, as well as the control sample.

Table 4 presents the results of the sensory evaluation of fried noodles with the additions of hemp protein and hemp protein with TGase. There were no significant differences in the assessment of their smell before cooking. The other quality features differed significantly. Despite the color and characteristic taste, the instant noodles with additives received a large number of points in the sensory evaluation. The control sample received the highest number of points. On the other hand, the lowest number of points for appearance before (4.2) and after cooking (4.3), as well as taste and smell after cooking (4.2), was obtained for instant noodles with 5% protein. With the increases in the levels of additives, instant noodles received fewer points for all quality characteristics. The addition of TGase resulted in better scores in the sensory evaluation.

Table 3. Average values of the results of the sensory analysis of instant noodles with the additions of rice protein, rice protein and TGase. Mean values denoted by different letters ([a]) represent statistically significant differences between variables ($p < 0.05$).

	Protein Content [%]	Parameter			
		Appearance before Cooking	Smell before Cooking	Appearance after Cooking	Smell and Taste after Cooking
Control sample	0	5.0 [a]	5.0 [a]	5.0 [a]	5.0 [a]
Rice protein	1	4.8 [a]	5.0 [a]	4.9 [a]	4.9 [a]
	2	4.9 [a]	4.9 [a]	4.9 [a]	5.0 [a]
	3	4.8 [a]	4.9 [a]	4.8 [a]	4.8 [a]
	4	4.7 [a]	4.8 [a]	4.8 [a]	4.8 [a]
	5	4.8 [a]	4.8 [a]	4.7 [a]	4.8 [a]
Rice protein and 1% TGase	1	4.9 [a]	5.0 [a]	4.9 [a]	5.0 [a]
	2	4.9 [a]	5.0 [a]	4.9 [a]	4.9 [a]
	3	4.8 [a]	4.9 [a]	4.9 [a]	5.0 [a]
	4	4.8 [a]	4.9 [a]	4.9 [a]	4.9 [a]
	5	4.8 [a]	4.9 [a]	4.8 [a]	4.9 [a]
Rice protein and 2% TGase	1	5.0 [a]	5.0 [a]	5.0 [a]	5.0 [a]
	2	5.0 [a]	4.9 [a]	5.0 [a]	5.0 [a]
	3	5.0 [a]	5.0 [a]	5.0 [a]	5.0 [a]
	4	5.0 [a]	4.9 [a]	5.0 [a]	4.9 [a]
	5	5.0 [a]	4.8 [a]	4.9 [a]	4.9 [a]

Table 4. Average values of the results of the sensory analysis of instant noodles with the addition of hemp protein and hemp protein and TGase. Mean values denoted by different letters ([a,b]) represent statistically significant differences between variables ($p < 0.05$).

	Protein Content [%]	Parameter			
		Appearance before Cooking	Smell before Cooking	Appearance after Cooking	Smell and Taste after Cooking
Control sample	0	4.8 [b]	4.9 [a]	4.8 [ab]	4.8 [ab]
Rice protein	1	4.7 [ab]	4.8 [a]	4.7 [ab]	4.7 [ab]
	2	4.7 [ab]	4.7 [a]	4.6 [ab]	4.6 [ab]
	3	4.6 [ab]	4.6 [a]	4.5 [ab]	4.4 [ab]
	4	4.2 [a]	4.5 [a]	4.3 [a]	4.2 [a]
	5	4.9 [b]	4.9 [a]	4.9 [ab]	4.8 [ab]
Rice protein and 1% TGase	1	4.9 [b]	4.9 [a]	4.8 [ab]	4.8 [ab]
	2	4.8 [b]	4.8 [a]	4.7 [ab]	4.7 [ab]
	3	4.8 [b]	4.8 [a]	4.6 [ab]	4.7 [ab]
	4	4.7 [ab]	4.8 [a]	4.5 [ab]	4.6 [ab]
	5	4.9 [b]	4.9 [a]	4.9 [ab]	4.9 [b]

Table 4. *Cont.*

	Protein Content [%]	Parameter			
		Appearance before Cooking	Smell before Cooking	Appearance after Cooking	Smell and Taste after Cooking
Rice protein and 2% TGase	1	4.9 [b]	4.8 [a]	4.9 [ab]	4.9 [b]
	2	4.9 [b]	4.8 [a]	4.8 [ab]	4.8 [ab]
	3	4.9 [b]	4.8 [a]	4.8 [ab]	4.8 [ab]
	4	4.9 [b]	4.8 [a]	4.7 [ab]	4.7 [ab]
	5	4.8 [b]	4.9 [a]	4.8 [ab]	4.8 [ab]

3.7. PCA

Principal component analysis (Figure 9) of the results of 31 evaluated instant fried noodle samples showed that sample variation corresponded to the first main component (Factor 1), which accounted for 46.58% of the total variability and was mainly related to Color components (L*, a*, b*), Hardness, Appearance after cooking, Smell and Taste after cooking and Small and Appearance before cooking. The second component (Factor 2) constituted 18.42% of the general variable and was mainly related to Fat content, Firmness, Water content and Appearance after cooking (Figure 4).

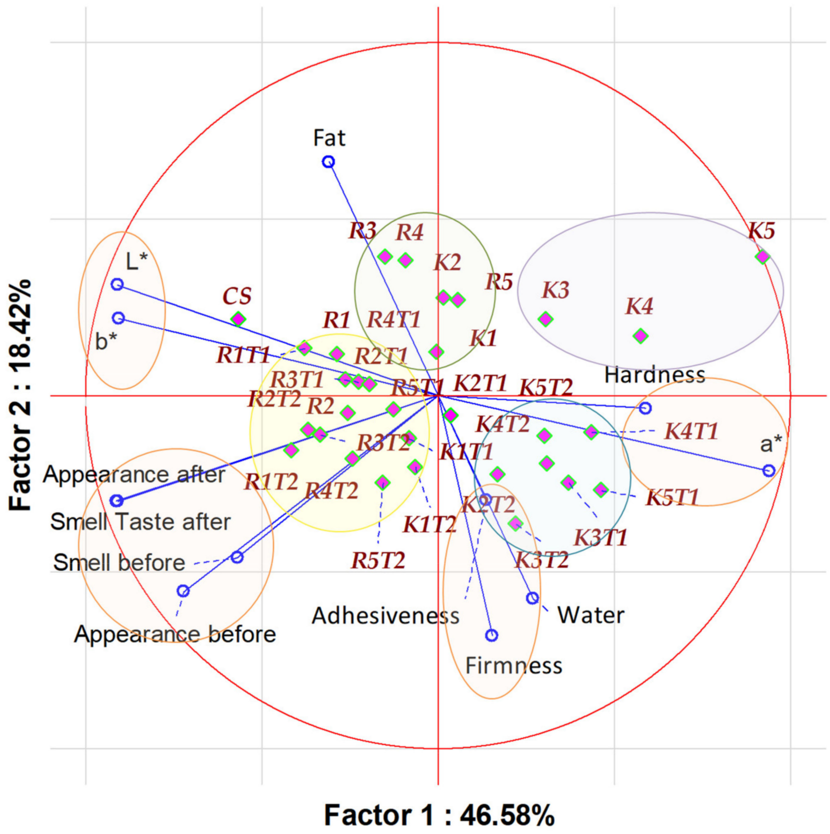

Figure 9. Principal component analysis (PCA) of the following samples: CS-control sample, R1–R5-CS with rice protein (from 1% to 5%, respectively), R1T1–R5T1-CS with 1% TGase and rice protein (from 1% to 5%, respectively), R1T2-R5T2-CS with 2% TGase and rice protein (from 1% to 5%, respectively), K1–K5-CS with hemp protein (from 1% to 5%, respectively), K1T1–K5T1-CS with 1% TGase and hemp protein (from 1% to 5%, respectively), K1T2–K5T2-CS with 2% TGase and hemp protein (from 1% to 5%, respectively). L*, b* and a* correspond to color components, 'before' and 'after' correspond to parameters measured before and after cooking.

The PCA results show that analyzed samples can be clustered into four distinctive groups. One of them consisted of noodle samples containing additions of hemp protein in amounts higher than 3%. The second cluster contained mostly noodles with various hemp protein additions with 1% TGase. The next cluster contained noodles with rice protein additions in amounts higher than 3% and instant noodles with low additions of hemp protein. The last cluster consisted of instant noodles with additions of rice protein and TGase in amounts of 1% and 2%.

The obtained clusters distinguished samples with different types of protein and amounts of TGase added. Instant noodle samples with the addition of rice protein without TGase were characterized by higher levels of fat, lower levels of water content, and higher level of firmness. Additionally, appearance and smell before and after cooking were not positively accepted and were not intense. The control sample was characterized by the highest level of color and highest level of sensory parameters among the analyzed noodles, but was also correlated with higher amounts of fat than other samples. Samples containing TGase were associated with a better appearance than samples without TGase, the level of fat was lower, and firmness as well as water content were higher in comparison to samples without TGase.

The addition of TGase together with rice protein resulted in better organoleptic features than other samples. In contrast, the addition of hemp proteins with 2% TGase resulted in better physicochemical properties, such as higher water content, firmness and adhesiveness. The addition of rice and hemp protein decreased the values of color components. Samples made with hemp protein were correlated with increased hardness levels: it was found that the higher the amount of hemp protein, the harder the analyzed sample. The addition of 2% TGase increased the level of hardness.

Based on the provided analysis, one can conclude that samples with different proteins and different levels of TGase added were characterized by different factors. The addition of protein increased the levels of L*, and b* components in comparison to the control sample, and the addition of hemp protein caused higher differences. On the other hand, the addition of proteins alone did not improve the level of fat content. Samples with rice protein and additions of 2% TGase were perceived to be better by the consumers, but lower levels of fat could be observed in samples with hemp protein and 2% TGase additions. The only problem was found with the color parameter analysis, because the samples with the best quality had behaviors far from corresponding to control sample. One can conclude that the obtained results suggested that the higher the level of TGase addition, the better the organoleptic and sensory features of instant fried noodles.

4. Conclusions

The presented analysis was devoted to determining the effects of rice and hemp protein additions together with transglutaminase on the quality characteristics of fried instant noodles, as well as decreasing the fat content.

The results obtained in our research confirmed that the addition of rice and hemp protein increased the water content. Instant noodles with the addition of TGase were characterized by the significantly lower fat content in comparison to the others. All instant noodles obtained during the research process were characterized by a short hydration time, not exceeding 5 min. The instant noodles with the addition of rice protein and the addition of TGase reduced the hardness. The additions of rice and hemp protein with the 2% addition of TGase increased the firmness of the instant noodles. The additives used impacted on the L*, a* and b* color parameters of the instant noodles. Along with the increase in the content of both rice protein and hemp protein, the value of the L* parameter decreased. All analyzed samples of the instant noodles obtained scores over 4.2 points on a five-point scale, and it can be observed that different combinations of additives used did not significantly reduce the sensory quality of instant noodles.

The most promising sample among all analyzed instant noodles was the sample with the addition of 5% rice protein with 2% TGase-a higher reduction in fat content (from 25% to 16%) with a high level of quality and acceptance level was observed.

The proposed research could be extended by a further study by improving the combination of additives, and by expanding the range of the analysis to improving the consumer acceptability level and to increase health values through reductions in the fat content.

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