Journal of Engineering Science *Vol. XXXIII, no. 1 (2026), pp. 146 - 164*
Fascicle *Food Engineering* *ISSN 2587-3474*
Topic *Food Technologies and Food Processes* *eISSN 2587-3482*

https://doi.org/10.52326/jes.utm.2026.33(1).10
UDC 664.681.15:664.64:543:635.658

PHYSICOCHEMICAL, SENSORY AND TEXTURAL EVALUATION OF SUGAR BISCUITS SUPPLEMENTED WITH LENTIL FLOUR

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Received: 01. 23. 2026
Accepted: 03. 12. 2026

Abstract: Grain legumes are widely acknowledged as fundamental sources of dietary protein on a global scale. In particular, lentil proteins contribute not only essential amino acids to the human diet but also serve as a source of bioactive peptides with documented health-promoting properties. The present study aimed to evaluate the impact of lentil flour (*Lens culinaris*) incorporation on the quality attributes, color, and textural properties of sugar biscuits. Lentils are highly valued due to their well-balanced nutritional composition. The analyzed red lentil variety exhibited a notable chemical profile, characterized by a protein content of 31.35 g/100 g, lipid content of 1.87 g/100 g, and dietary fiber content of 12.6 g/100 g. Sugar biscuits formulated with varying levels of lentil flour (1–15%) were assessed in terms of sensory characteristics, physicochemical properties, texture parameters, and color indices. Among the tested formulations, the sample containing 10% lentil flour achieved the highest overall evaluation score. Based on the obtained quality indicators, the biscuits can be considered safe for consumption, maintaining their key quality attributes over a storage period of up to 35 days under specified conditions. Consequently, these products may be recommended as part of the diet for diverse consumer groups.

Keywords: *legumes, lentil, flour, biscuits, texture, sensory quality, physicochemical quality.*

Rezumat. Boabele leguminoaselor sunt recunoscute pe scară largă ca surse importante de proteine alimentare la nivel global. În mod particular, proteinele din linte furnizează nu doar aminoacizi esențiali, ci constituie și o sursă de peptide bioactive cu efecte benefice asupra sănătății. Scopul prezentului studiu a fost evaluarea efectului încorporării făinii de linte (*Lens culinaris*) asupra caracteristicilor de calitate, culorii și proprietăților texturale ale biscuiților zaharoși. Lintea este apreciată pentru compoziția sa nutrițională echilibrată. Soiul de linte roșie analizat a evidențiat un profil chimic valoros, caracterizat printr-un conținut de proteine de 31,35 g/100 g, lipide de 1,87 g/100 g și fibre alimentare de 12,6 g/100 g. Biscuiții zaharoși formulați cu diferite proporții de făină de linte (1–15%) au fost evaluați din punct de vedere al caracteristicilor senzoriale, proprietăților fizico-chimice, parametrilor texturali și indicilor de culoare. Dintre variantele analizate, proba cu adaos de 10% făină de linte a înregistrat cel mai ridicat scor global. Pe baza indicatorilor de calitate determinați, biscuiții pot fi considerați siguri pentru consum, menținându-și caracteristicile esențiale pe o perioadă de depozitare de până la 35 de zile, în condiții specificate. În consecință, aceștia pot fi recomandați pentru includerea în alimentația diferitelor categorii de consumatori.

Cuvinte cheie: *leguminoase, linte, făină, biscuiți, textură, calitate senzorială, calitate fizico-chimică.*

1. Introduction

The active and healthy lifestyle of people requires a rational diet presented by the content of essential amino acids in food, which are involved in metabolic processes, energy generation and regeneration. The vegetarian diet does not accept animal products. Protein deficient diet can lead to many disorders such as colon cancer, heart disease and osteoporosis [1, 2]. Thus, for a complete existence, it is necessary to find ways to enrich the daily diet with protein products.

Lentils (lat. *Lents*) is a leguminous plant native to the Near East and is one of the oldest legumes cultivated alongside wheat, barley and peas. Lentils have a well-balanced nutritional profile, consisting of 22.7% protein (including all essential amino acids), 51% carbohydrates, 13.8% dietary fiber, and 1% lipids [3]. Lentils are a valuable plant-based protein-rich product [4].

Due to its nutritional value, the possibility of using lentils, namely, lentil flour as a component of the recipe for the preparation of floury confectionery products, is of scientific interest. Wilson et al. [5] concluded that lentil consumption improved insulin sensitivity in obese individuals, contributing to lower blood glucose and improved insulin status. In other works, it has been reported [6, 7, 8] that lentil consumption improves arterial elasticity, counteracts atherosclerotic disease and helps lower blood pressure. Other researchers have reported that the consumption of lentils may have a dietary effect for people facing obesity as a preventive measure against cancer, chronic diseases and hypertension during life [1, 9]. In recent years, lentils have become a popular food among people who suffer from a genetic dysfunction and cannot digest some proteins in the cereal composition (gluten intolerance) [10].

Biscuits are a popular bakery product, but their low content of protein, fiber, minerals, and vitamins makes them less suitable for regular daily consumption [11]. Considering that biscuits are flour confectionery products in constant demand [11] by the population, especially children, adolescents and the elderly, there is a great interest in their fortification with value-added compounds [10, 12-14]. Biscuits occupy the first place (36%) in the Republic of Moldova in terms of the flour confectionery products volume (Figure 1).

The mixture of flours containing legumes and wheat flour could effectively increase the nutritional and functional value of the product [15]. Fortification with food ingredients rich in proteins and micronutrients is one of the main strategies adopted to improve the nutritional quality of the population in rural areas of developing countries [16]. For young children, fortified crackers are widely used as a way to solve the crucial problem of malnutrition. The enrichment of products with vegetable proteins can be achieved by introducing legumes into the technological recipes of flour confectionery products. This highlights the potential of legumes to enrich the product with biologically active substances, to increase its nutritional and biological values, to increase the yield of the finished product and to contribute to the increasing the shelf life of food products [17, 18].

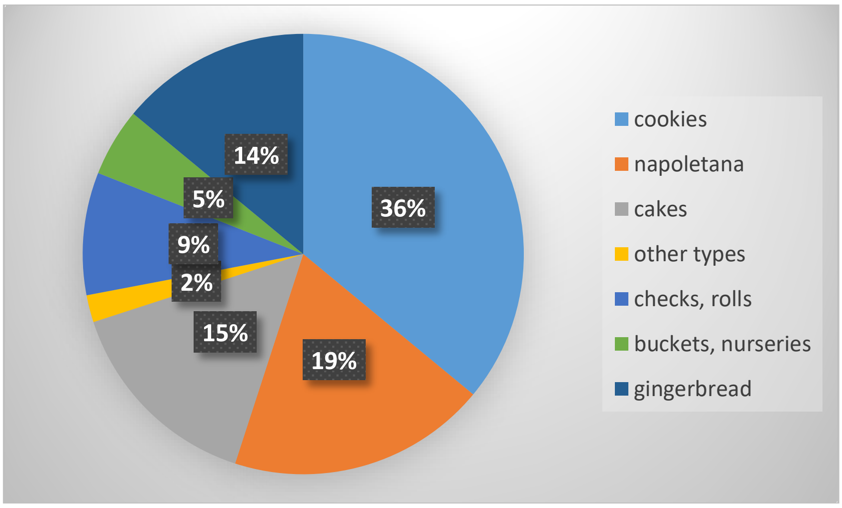

Figure 1. Structure of the manufacturing flour confectionery products volume [1].

According to the results of other scientists, the addition of lentil flour has been proposed to enrich with dietary fiber up to 20%, which will help to increase the protein content of the product up to 2 – 5%, also in order to reducing the energy value of the product [17-20].

The quality of flour confectionery products is a combination of properties that determine their ability to rationally satisfy human needs for a healthy diet. The nutritional value of flour confectionery products is determined by the quantitative content and qualitative composition of proteins, fats, carbohydrates, minerals, vitamins, as well as their energy value [4].

The aim of the work was to develop a technology for manufacturing sugary biscuits with lentil flour and to investigate its effect on the quality of the end product, color parameters, textural properties and microbiological stability during storage.

2. Materials and Methods

2.1. Chemical Materials

Hydrochloric acid (37 %), sodium hydroxide (97 %), phenolphthalein, bromothymol blue, sodium nitrite (≥97,0 %), aluminum chloride, sodium citrate (≥99,0 %), trisodium citrate, silver nitrate (≥99,0 %), hexane reagent (>99 %), hydrochloric acid (37 %), KjTabs VST, Vreceiver TKN, sulfuric acid (95,0–98,0 %), sodium hydroxide (≥85 %), were purchased from Sigma (Darmstadt, Germany; Tokyo, Japan; Shanghai, China).

2.2. Biological Materials

The seeds of red lentils *(Lens culinaris* L.*)*, peas *(Pisum sativum)* and beans *(Phaseolus vulgaris)* were received from the Institute of Genetics, Physiology and Plant Protection, Chisinau.

2.3. Characteristic of Wheat and Legume Flour

Legume seeds were dried in a laboratory dryer (SLW 115 SMART) with forced air circulation at a constant temperature of 50 ± 2 °C and relative air humidity of 60–65 %. The drying process was carried out for 5 hours. The final moisture content of the dry seeds was 14.02± 0.04%. Legume seeds were crushed in a laboratory hammer mill (PERTEN LM 120, Sweden) into powder with a particle size of 180±15 μm, sieved and vacuum packed for further use in this study.

2.3.1. Sensory Analysis of Wheat Flour and Red Lentil Flour

Sensory indices were determined for wheat and lentil flour in terms of pest infestation, colour, odor and taste according to BS ISO 6658-2005 – Sensory Analysis – Methodology – General quidance [21].

2.3.2. Physicochemical Analysis of Wheat and legumes

The moisture (method 44-19), ash (method 08-01), crude protein (method 46-12), crude fat (method 30-25), and total fiber contents were determined according to [22]. Fat content was quantified gravimetrically following hexane extraction from the dehydrated sample using the Soxhlet method with a SER148 Solvent Extraction Unit (VELP Scientifica, Monza, Italy). Protein content was estimated by determining total nitrogen and applying a conversion factor of 6.25, using the Kjeldahl method with a UDK129 unit (VELP Scientifica, Italy). Titratable acidity was measured by titrating a known volume of sample with standard 0.1 N NaOH, using phenolphthalein as an indicator [23]. The pH was measured using a pH meter (TESTO 206-pH2, Pruszków, Poland), calibrated with pH 4.0 and 7.0 buffer solutions. The electrode was directly immersed in a beaker containing the sample macerated with distilled water, following the method [24].

Wet gluten content [25 - 27] was carried out by washing the dough prepared from the analyzed flour, the water temperature being 18-20°C and whipping the obtained gluten.

Equation (2) was used to determinate the wet gluten content, expressed in %:

$$Wet\ gluten = \frac{m_1}{m} \cdot 100, \qquad (1)$$

where:

m_1 – wet gluten mass remaining after churning, g;

m – flour sample mass taken for determination, g.

Gluten quality: elasticity was achieved using the IDK-3M device by determining the ability of gluten to resist compression [25-27]. The extensibility consists in manually stretching the wet gluten (5 g) modeled in the form of a wick, until breaking, under established conditions and measuring the length reached by the gluten at the time of breaking [25-27].

2.4. Preparation and Characterization of Sugar Cookies with the Addition of Red Lentil Flour

2.4.1. Preparation of Sugar Cookies with the Addition of Red Lentil Flour

Biscuits samples were made using 1% to 15% lentil flour (SLF1 – sugary biscuits with 1% lentil flour; SLF5 – sugary biscuits with 5% lentil flour; SLF 10 – sugary biscuits with 10% lentil flour; SLF 15 – sugary biscuits with 15% lentil flour.). For the control, a sample was prepared without the addition of LF (CS – control sample,100% high quality wheat flour). High quality wheat flour, sugar, butter, salt and baking powder were used to make sugar cookies. Respective raw materials were purchased from the local market. Sample recipes were calculated according to the dry matter and moisture content of the raw material, Table 1.

To prepare the dough, a mixture of sugar and fat is made, homogenized for 3-4 minutes, after which approximately 50% of the wheat flour is added. Knead in the bowl of the food processor (Tefal QB150138, Germany) for 7-10 min, then salt, the rest of the wheat flour and lentil flour, chemical softener, vanillin were added and kneaded for about 10 min.

Table 1

Recipe of experimental samples

Components	CS	SLF 1	SLF 5	SLF 10	SLF 15
Premium quality wheat flour, g	100	99	95	90	85
Red lentil flour, g	0	1	5	10	15
Cow butter,g	25	25	25	25	25
Salt, g	2	2	2	2	2
Powdered sugar, g	35	35	35	35	35
Chemical softener, g	2	2	2	2	2
Vanilla, g	0,7	0,7	0,7	0,7	0,7

Note: CS – control sample, 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour.

The temperature of the kneaded dough was 23±1 °C. The rested dough was subjected to lamination in the form of a continuous sheet of 2-4 mm thickness. The modeling was done by hand using the die that cuts the modeled pieces. The stamped blank with a mass of 15 ± 1 g was baked in an electric convection oven (EASY EV-UME604-LS, Luxstahl, PiggioTorriana, Italy) at 190 ± 2 °C for 10 ± 1 min. After baking, the biscuits were cooled, packaged and stored in a dry place at room temperature.

2.4.2. Physicochemical Analysis of Dough and Sugar Biscuits

The moisture content of dough and sugar cookies was determined according to the AACC standard (AACC, Method 44-15.02) [28]. Alkalinity was determined by neutralization by titration with 0.1 N hydrochloric acid solution in the presence of bromothymol blue as an indicator and is expressed in degrees of alkalinity [29].

The absorption index of sugar cookies was determined according to [30].

The water activity (a_w) of the baked biscuits was measured at room temperature (20 ± 1 °C) using the LabMaster dew point water activity apparatus (Novasina AG, CH-8853 Lachen, Switzerland) [31]. The determinations were carried out within 35 days: on the 1st day, the 10th, the 20th and the 35th day.

2.4.3. Texture Profile Analysis of Dough

A Stable Micro Systems TA.HD Plus C analyzer (UK) was used to perform texture profile analysis of sugar biscuits dough. The textural properties of the dough samples—including hardness, cohesiveness, gumminess, resilience, and adhesiveness—were determined using a 40 mm cylindrical probe through a double compression test with a P/75 stainless steel plate. The test was conducted under the following conditions: pre-test speed of 100 mm/s, test speed of 5 mm/s, post-test speed of 5 mm/s, and a load cell capacity of 5 kg [32]. Textural determinations were performed on the day of dough preparation.

2.4.4. Sensory Analysis of Sugar Cookies

The sensory analysis of the biscuits was conducted by a panel of nine assessors, who evaluated the following attributes: shape, surface, color, taste, aroma, and cross-sectional appearance. Sensory quality was assessed using a 5-point scale. The evaluation was performed in individual booths under white light at room temperature. The panel developed a set of attributes and scored the samples as follows: 5 – very good (exceptional quality, ideal); 4 – good (acceptable quality); 3 – satisfactory (minor defects); 2 – unsatisfactory (clearly noticeable defects); 1 – poor (pronounced defects); 0 – very poor (major deterioration). Each sample was analyzed in parallel in a laboratory designed for sensory evaluation in accordance with ISO 8589 [33]. The samples were served in lidded plastic containers, coded with random numbers, and presented in random order. A break of up to 5 minutes was allowed between evaluations. The determinations were carried out the following day.

2.4.5. Microbiological Analysis of Sugar Biscuits

The microbiological investigation: the dilution method was used for the quantitative calculation of colony forming units (CFU) in determining the corresponding groups of microorganisms in 1 cm^3 of bacterial solution. Basic dilutions (10^1, 10^2, 10^3) were prepared as follows: 1 g of each biscuit sample was added to a test tube containing 9 cm^3 of saline (0.85%) or sterilized water. In Petri dishes with a gel-like nutrient substrate, 1 cm^3 of a bacterial solution was inoculated in triplicate. For the cultivation of microorganisms, two types of nutrient media were used to separate groups of microorganisms: agar for bacteria (48 h of incubation at 28 °C) and Sabouraud for yeast (5 days of incubation at 25 °C). After incubation, the results were analyzed and different types of microorganisms were identified in terms of cultural and morphological characteristics. The bacterial culture was further identified by Gram staining [34-37]. The determinations were carried out within 35 days: on the 1st day, the 10th, the 20th and the 35th day.

2.4.6. The Energy Value of Sugar Cookies

The energy value of sugar cookies (kcal/100g) was calculated. The conversion factors for calculating the energy value of the main nutrients in food were as follows, in kcal/g: carbohydrates – 4; proteins – 4; lipids (fats) – 9; fibers – 2 [38].

2.5. Color Analysis of Lentil Flour and Sugar Biscuits Using the CIELAB Method

The CIELab parameters of sugar cookies with the addition of lentil flour were measured using a Chroma Meter CR-400/410 colorimeter (Konica Minolta, Tokyo, Japan), according to the method [39]. Each sample was analyzed at three distinct points, measuring the variations in luminance (L*), red/green component (a*), yellow/blue component (b*). The overall color difference (ΔE*) was calculated according to Eq. (2).

$$\Delta E^* = \sqrt{(L_i^* - L_0^*)^2 + (a_i^* - a_0^*)^2 + (b_i^* - b_0^*)^2}, \quad (2)$$

where:

L_0^*, a_0^* and b_0^* – the sample values on the day of baking;

L_i^*, a_i^* and b_i^* –the sample values in storage days.

The determinations were carried out within 35 days: on the 1st day, the 10th, the 20th and the 35th day.

2.6. Statistical Analysis

All analyses were performed in triplicate, and the results are expressed as mean values ± standard error of the mean. Microsoft Office Excel 2007 (Microsoft, Redmond, WA, USA) was used for data processing. Statistical analysis was conducted using one-way analysis of variance (ANOVA), followed by Tukey's test at a significance level of $p \leqslant 0.05$, using Statgraphics Centurion XVI version 16.1.17 (Statgraphics Technologies, Inc., The Plains, VA, USA).

3. Results and Discussion

3.1. Characteristic of Wheat Flour and Legumes

The vegetable raw material from the Leguminosae family *(Fabaceae or Papilionaceae family)* – lentils, beans and peas – were analyzed. Lentil seeds have a high content of protein and dietary fiber compared to peas and beans, this can be seen from Table 2.

According to other researchers, lentils are rich in protein and these values vary between 22-31% [40 -42], the protein content of peas [43] varies between 21-27% and beans 20 -41% [40, 44]. The protein content obtained from the research should fall within these values, being 31.35±0.79, 23.01±0.01 and 21.04±0.12, respectively, for lentils, peas and beans.

Table 2

The nutritional value of legumes

Legumes	Protein content, %/100g	Fat content, %/100g	Carbohydrate content, %/100g	Dietary fiber content, %/100g	Moisture content, %	Ash, %	Energy value, kcal/100g
Lentils	31.35± 0.19 c	1.87± 0.01 b	43.30 ± 0.12 a	12.60± 0.15 c	11.27± 0.05 c	2.33± 0.01 a	314
Peas	23.01± 0.24 b	1.60± 0.02 a	48.00± 0.19 c	10.70± 0.09 b	9.71± 0.10 a	3.45± 0.03 b	317
Beans	21.04± 0.12 a	2.00± 0.02 b,c	47.00± 0.13 b	10.40± 0.16 a	10.86± 0.08 b	3.50± 0.05 b	286

Note: Different letters (a-c) designate statistically different results ($p \leqslant 0.05$).

Grusak [45] reported the following data on fat content, compiled from different studies: for lentils – 0.3-3.5% [40 - 42], for peas –1.2 – 7.3% [46, 47], for beans – 1 .66 – 2.13% [48].

The values of moisture content 9.71±0.02% for peas, 10.86±0.02% for beans and 11.27±0.01 for lentils are similar to the study data [46, 49].

The results regarding the dietary fiber content of legumes varied from 10.4±0.02 to 12.6±0.05%. Lentils were the richest source of fiber, followed by peas and beans. The data were in agreement with [46, 47, 50] who reported that dietary fiber in lentils is found to be 7.9-12.0%, in peas and beans, 16.7 % and 10-20%, respectively [47, 50, 51]. The ash content in the investigated samples was 2.33±0.01 for lentils, in peas 3.45±0.01 and in beans 3.50±0.01%.

Table 2 shows that lentils contain the highest amount of protein and fiber (24% and 12.5% respectively) compared to peas and beans. It is lower in carbohydrates and lipids (46% and 1.5% respectively).

For the manufacture of biscuits, the quality of wheat flour and lentil flour (LF), which correspond to the norms provided in the normative acts, was analyzed sensoryally, Table 3.

Table 3

Organoleptic analysis of wheat and lentil flour

Flour type	Organoleptic indices			
	Color	Odor	Taste	Pest infestation
Premium quality wheat flour	White with a slight yellowish tint	Pleasant, characteristic of flour, without foreign or musty smell	Sweet, without crunch when chewing	Absent

Continuation Table 3

Lentil flour	Yellow characteristic of the variety	Pleasant, characteristic of flour, without foreign or musty smell	Characteristic, without crunch when chewing	Absent

According to the organoleptic indicators, the flours should be certified according to the color characteristic of the variety, with a pleasant taste and smell without the smell of foreignness or mold. The data obtained regarding the organoleptic analysis of high-quality wheat and lentil flour are correlated with the data of the normative acts in force in this field [52, 53].

The physicochemical indicators of wheat and LF were determined and are presented in Table 4.

Table 4

Physico-chemical indicators of wheat and lentil flours

Indicators	Premium quality wheat flour	Lentil flour
Moisture content, %	13.50±0.08	11.27±0.01
Acidity, degree	2.65±0.04	4.05±0.02
Ash, %	0.47±0.02	2.33±0.01
Gluten content, %	22.00±0.01	Not determined
Elasticity, c.u.	69.0±0.1	Not determined
Extensibility, cm	14.0±0.1	Not determined
Fat content, g/100g	1.00±0.02	1.87±0.01
Protein content, g/100g	10.3±0.02	31.35±0.19
Total fiber content, g/100g	2.68±0.02	12.6±0.15
Finesse, %	Characteristic of wheat flour	Characteristic of lentil flour
The content of ferromagnetic impurities, mg/kg	Not detected	Not determined

The obtained data regarding the analysis of the physico-chemical indicators of high-quality wheat flour are correlated with the data of the in force normative acts [52, 53].

The moisture content of lentil flour was lower by 1.2% compared to wheat flour, similar results being obtained by other authors [54]. The low moisture content of lentil flour protects it from the danger of mold growth. The acidity of lentil flour (4.05 degree) was higher than the acidity of wheat flour (2.65 degree). This is probably related to the presence of free fatty acids in lentils [55]. Lentil flour was characterized by a major content of mineral substances [56, 57]. The protein content of lentils was high and varies between 19.5% - 35.5% [40]. Lentil proteins were highly valued due to their high digestibility (~83 %) [58]. The analyzed red lentil variety presented an appreciable chemical composition, with a protein content of 31.35 g/100g. The fiber content of lentil flour was 5 times higher than that of wheat flour [3].

3.2. Characteristic of dough and sugar cookies

The dough was prepared according to the established recipe and subjected to the technology described in p. 2.4.1. The determinations were carried out the following day. The results obtained, regarding the physico-chemical evaluation of the dough, are presented in Table 5.

Table 5

Physico-chemical characteristics of dough with red lentil flour

Sample	Characteristics	
	Moisture content, %	Alcalinity, degrees
CS	27.0±0.3 [a]	1.00±0.01 [e]
SLF 1	28.3±0.5 [b]	0.96±0.01 [d]
SLF 5	29.7±0.4 [c]	0.92±0.01 [c]
SLF 10	30.2±0.5 [c,d]	0.88±0.0 [b]
SLF 15	32.12±0.2 [d]	0.84±0.01 [a]

Note: CS – control sample – 100 % Premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour. Different letters ([a–e]) designate statistically different results ($p \leq 0.05$).

Analyzing the experimental results, an increase in moisture values was observed from 27.00% (CS) to 32.12% (SLF 15), by 1.19 times. This contributed to the increase the dough's hydration capacity. This was explained by the fact that the higher the protein and soluble fiber content, the higher the flour hydration capacity. According to [59] the dough could become softer due to the increased solubility of sugar in the dough formulation. Similar results were obtained by Taylor et al. [60], who stated that sugars with a higher solubility result in a soft dough. Dough alkalinity decreased with increasing lentil flour addition due to more organic acids and alkaline substances in lentil flour compared to wheat flour, such as potassium and magnesium. The alkalinity indicator did not exceed the conditions regarding admissibility (at most 2.0 degrees) [61].

During the dough kneading process, the dough was observed to have superior structural-mechanical properties, which aided kneading and was explained by the lipid content of the lentil flour [62].

Dough is a material with visco-elastic properties due to the presence of glutenins and gliadins in its molecular structure. Dough texture is not usually measured during mixing and processing, but its rheological properties are used as a guide for dough processing and its influence on the baked product structure, being even an indicator of the appearance and quality of the finished food products [63]. In the further processing of the dough after mixing, its relaxation played an important role on the dough textural characteristics [64]. The texture parameters of the dough with the addition of LF were presented in Table 6.

Table 6

Texture parameters of sugar biscuits dough with the addition of lentil flour (results are presented as mean ± standard deviation)

Dough samples	Hardness, g	Cohesiviness, %	Resilience, %	Gumminess, g	Chewiness, g
CS	1146.67±40.07 [a]	0.526±0.006 [d]	0.155±0.001 [d]	603.14±10.20 [a]	603.75±10.02 [a]
SLF 1	1301.88±20.09 [b]	0.518±0.009 [c,d]	0.149 ±0.002 [c]	674.37±15.16 [b]	675.04±13.11 [b]
SLF 5	1576.49±18.05 [c]	0.504±0.010 [c]	0.147±0.001 [b,c]	794.55±10.11 [c]	795.34±9.07 [c]
SLF 10	1863.90±32.11 [d]	0.473±0.006 [b]	0.137±0.001 [a]	881.62±13.26 [d]	883.38±12.09 [d]
SLF 15	1981.11±25.07 [e]	0.440±0.006 [a]	0.135±0.001 [a]	871.68±5.20 [d]	873.42±8.16 [d]

Note: CS – control sample – 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour. Different letters ([a–e]) designate statistically different results ($p \leq 0.05$).

Dough hardness increases for all samples with values between 11.92% and 42.11%. The hardness of the dough increased because of the addition of lentil flour, which is a gluten-free raw material that could contribut to the reducing the gluten amount and could have a negative effect on its structure. Similar results were obtained by the authors [50.] when they substituted wheat flour with 10 % germinated lentil flour to obtain bread, where the increase in hardness was about of 14 %. Also, the increase in the hardness value can be affected by the interactions between the wheat flour gluten and the lentil flour dietary, fibers with increased water retention capacity [65]. This combination can weaken the gluten structure. On the other hand, the increase in dough hardness can be influenced by the interaction of starch with the increased amount of protein, due to the addition of lentil flour, which is a protein rich legume [66].

The obtained results for cohesiveness and resilience were decreasing, proportionally with the increase in the content of lentil flour in the dough, by 19.50 % and almost 15 %, respectively. As in the case of hardness, the values for these two parameters are mainly affected by the decrease in the gluten amount of the end product, considering the wheat flour replacement with lentil flour. Similar results were obtained by other authors [67] where they used germinated lentil flour to replace wheat flour, and the decrease in the respective parameters values was almost 3 times compared to the control sample. The values for gumminess and chewiness are higher compared to the control sample. For both parameters, the highest increase, 31 %, was observed for the dough with the addition of 10 % lentil flour. According to other authors [68] the obtained results are characteristic for more compact structures that could be due to a higher content of food fibers in the end product, coming from the added lentil flour and the changes that the starch undergoes. The values for these parameters increased proportionally with the increase in dough of the lentil flour content.

3.3. Characteristic of biscuits

One of the marketing requirements for biscuits is to increase their shelf life. The quality of the biscuits was evaluated during the 35-day storage period, with the exception of the sensory analysis that was done on the day the biscuits were baked. The results of sensory and physico-chemical analysis of sugar cookies samples are presented in Table 7.

Table 7

Sensory analysis and energy value of sugar biscuits with lentil flour (results are presented as mean ± standard deviation)

Characteristics	Sugar biscuits				
	CS	**SLF 1**	**SLF 5**	**SLF 10**	**SLF 15**
Average score of sensory profile	4.59 ± 0.01 [a]	4.66 ± 0.01 [b,c]	4.90 ± 0.01 [d]	4.55 ± 0.01 [a]	4.50 ± 0.01 [a]
Consistency	4.81 ± 0.01 [b]	4.82 ± 0.02 [b,c]	4.89 ± 0.02 [c]	4.53 ± 0.02 [a]	4.51 ± 0.01 [a]
Taste	4.75 ± 0.02 [b]	4.81 ± 0.01 [c]	4.91 ± 0.01 [d]	4.75 ± 0.01 [b]	4.65 ± 0.01 [a]
Appearance	4.51 ± 0.01 [a]	4.65 ± 0.0 [b]	4.95 ± 0.02 [c]	4.65 ± 0.0 [b]	4.62 ± 0.02 [b]
Color	4.32 ± 0.01 [a]	4.51 ± 0.02 [b]	4.85 ± 0.01 [c]	4.50 ± 0.02 [b]	4.51 ± 0.01 [b]
Odor	4.56 ± 0.02 [d]	4.52 ± 0.01 [c,d]	4.91 ± 0.02 [e]	4.31 ± 0.01 [b]	4.21 ± 0.01 [a]
The energy value, kcal/100 g	368	367	364	361	357

Note: CS – control sample – 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour; L *—luminosity; a *—red/green component; b *—yellow/blue component; ΔE * - overall difference of color. Different letters ($^{a-l}$) designate statistically different results ($p \leq 0.05$).

Sensory analysis of the biscuits showed that the highest average scores were obtained for the SLF 10 sample (4.9 points), followed by the 15% PSC sample (4.55 points) and the control sample (4.59 points) (Table 9). The highest score for consistency was recorded for the

SLF 10 sample (4.89 points). Substituting 15% of wheat flour with lentil flour slightly reduced the score to 4.53 points, resulting in a more brittle texture. In contrast, the control sample and those containing 1−10% lentil flour exhibited a firm consistency. According to Idowu et al. [69], the firmer texture observed in biscuits containing 1−10% lentil flour may be attributed to amylose and amylopectin recrystallization, the formation of starch−protein complexes, and water redistribution among product components. All analyzed biscuit samples exhibited a pleasant and uniform external appearance. The incorporation of lentil flour into the biscuit formulation significantly improved the taste, aroma, and color, as noted by all panelists. Similarly, Saleem et al. [70] reported high overall acceptability for biscuits made with a wheat-to-lentil flour ratio of 93:7. Awadelkareem et al. [71] also observed high acceptability for biscuits containing 5% and 10% lentil flour. Hajas et al. [72] found that biscuits made with red lentils were more preferred than those made with black lentils. Good overall acceptability has also been reported for biscuits containing 10% lentil flour [73]. However, the addition of 15% SLF negatively affected the taste and aroma of the biscuits due to a pronounced lentil flavor.

An important factor in calculating the energy value is the fiber content, which is involved in digestion but does not provide calories. Dietary fiber is often included in energy value calculations because it is hypothesized that it may have an effect on calorie absorption [74, 75]. LF generally contains fewer calories than wheat flour. Replacing some or all of the wheat flour with lentil flour can reduce the calorie content of the biscuits. LF is high in protein, which can increase the nutritional value of biscuits, providing an additional source of protein for consumers, and it also contains a significant amount of fiber, which can improve digestion and gut health. This can also influence the level of energy gradually released during digestion, which can have an impact on the perceived energy value of the biscuits. LF can also contain a variety of essential vitamins and minerals, such as iron, zinc and B-complex vitamins, which contribute to the overall nutritional value of the bistuits. Overall, the use of LF in biscuits can provide significant nutritional benefits, such as increased protein and fiber intake, but can also influence energy value by reducing calories and managing energy release during digestion.

In order to maintain a healthy lifestyle and monitor the intake of calories and nutrients, the energy value of the experimental samples was calculated. The energy value of the product is the amount of energy that a food product provides upon consumption, measured in kilocalories per 100 g (kcal/100g) [76]. The nutritional information was consulted and the energy value resulting from the content of carbohydrates, fats and proteins, dietary fibers contained in the product was calculated [77]. The energy value of the biscuit samples is lower than the control sample. The energy value of the experimental samples is lower by 0.03% at the 1% LF experimental sample, by 1.1% at the 5% LF experimental sample, by 2.0% at the 10% LF experimental sample and by 3,0 % in the experimental sample of 15 % LF. As LF was added, the energy value decreased, because with the increase in the content of lentil flour added to the samples, the content of dietary fibers increases, which contributes to the nutritional and biological value of the product, but although they are involved in the process of digestion, they do not provide calories, so they do not influence the energy value. The decrease in the energy value of biscuits with added LF could be attributed to several factors. When LF is incorporated into cookie recipes, the overall composition changes. LF has a lower fat content compared to traditional wheat flour. In addition, LF is high in protein and dietary fiber, which can affect overall energy density. Protein and fiber provide satiety without contributing significantly to energy content [78.].

According to the results shown in Table 8, with the addition of lentil flour, the moisture values increase insignificantly, since the difference in moisture between lentil flour and wheat flour was 0.23%.

Table 8

Physicochemical changes in sugar biscuits enriched with lentil flour during storage (results are presented as mean ± standard deviation)

Indicators	Storage time, days	Sugar biscuits				
		CS	SLF 1	SLF 5	SLF 10	SLF 15
Moisture content, %	1	6.00±0.04 a,b	6.21±0.02 c	6.23±0.01 c	7.25±0.02 f	7.35±0.06 f,g
	10	6.01±0.01 a	6.31±0.02 d	6.32±0.02 d	7.35±0.03 g	7.37±0.05 g
	20	6.04±0.01 b	6.37±0.01 d	6.39±0.02 d	7.46±0.01 h	7.47±0.04 h
	35	6.05±0.02 b	6.40 ±0.05 d,e	6.49±0.02 e	7.58±0.02 h,i	7.61±0.06 h,i
Alcalinity, degrees	1	0.50±0.0 j	0.49±0.01 i,j	0.48±0.01 h,i	0.47±0.01 h,i	0.42±0.0 e
	10	0.46±0.02 g,h	0.45±0.01 f,g	0.45±0.01 f,g	0.44±0.01 f,g	0.41±0.01 e,d
	20	0.44±0.01 f,g	0.42±0.0 e	0.40±0.0 d	0.39±0.01 c,d	0.38±0.01 c
	35	0.40±0.0 d	0.38±0.01 c	0.36±0.0 b	0.35±0.0 b	0.33±0.0 a
Swelling index, %	1	127±1 e,f	125±2 d,e	118±2 b,c	117±1 b	115±1 a,b
	10	129±1 f,g	136±0 h	119±2 c	125±1 d,e	116±1 a,b
	20	138±1 h,i	139±1 h,i	124±2 d,e	132±0 g	119±1 b,c
	35	146±1 k	144±1 j,k	142±1 i,j	143±1 j	136±0 h
Water activity, a_w	1	0.384±0.001 e,f	0.417±0.001 h	0.413±0.002 h	0.411±0.002 h	0.401±0.002 g
	10	0.382±0.001 e	0.380±0.001 e	0.374±0.001 e	0.409±0.001 h	0.399±0.001 g
	20	0.327±0.0 d	0.326±0.001 d	0.324±0.002 d	0.312±0.001 c	0.311±0.001 c
	35	0.320±0.001 d	0.305±0.002 b,c	0.301±0.001 b	0.301±0.002 b	0.278±0.001 a

Note: CS – control sample – 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour; L *—luminosity; a *—red/green component; b *—yellow/blue component; ΔE * - overall difference of color. Different letters ($^{a–l}$) designate statistically different results ($p \leqslant 0.05$).

A slight increase in humidity can also be explained by the addition of lentil flour which contains proteins that intersect with wheat flour proteins, forming a protein network [79-81] and affecting the viscoelastic properties of the dough, leading to changes in the microstructure of the baked biscuits. It can influence texture, moisture retention [82]. The moisture content of all samples remains within the limits of the values indicated in the normative documents.

The alkalinity values obtained were below the permissible limit, not exceeding the standard specified in regulatory guidelines (<2.0 degrees) [3]. The slight decrease in the alkalinity values was probably related to the presence of free fatty acids in the chemical composition of the lentil flour, which was demonstrated by the titratable acidity value (4.05 degrees).

Quality biscuits must absorb water quickly and in large quantities. During storage, the control sample showed the best result, which was within the allowed values of at least 140%. In the experimental samples of biscuits with the addition of lentil flour, the soaking index values decreased. During storage for 35 days, this parameter depending on the addition of lentil flour decreased by 1.11 times in the experimental samples in the first 10 days, by 1.16 times – on the 20th day of storage, 1.2 times, on the 35th day - by 1.2 times. This is due to the structural characteristics of the biscuits: the high moisture content of the biscuit may indicate the presence of moisture in its structure [83], the use of certain ingredients such as sugar and butter, which may affect the texture and moisture content of the biscuit, resulting in a softer texture and higher moisture content [84], the low soaking index may be due to the denser structure of the biscuits, which makes it difficult for water to penetrate inside. Mohammadi et al. suggest that this effect can be attributed to the high protein and fiber content, as well as the presence of hydroxyl groups, which enhance the interaction with water molecules, thereby increasing moisture content and reducing the soaking index [85]. According to [86] the increase in moisture is due to the active increase in bound water as a consequence of the chemical and physical changes of the main components in the product.

The a_w results of the experimental samples showed that, with the increase in the amount of lentil flour, the water activity decreased. The obtained values indicated that in this product there was no probability of microbial development and product decomposition processes [87, 88]. The specialized literature attests that water activity values lower than 0.60 prevent microbial damage and show microbiological stability [89, 90].

The influence of storage time (35 days) on the CIELab color parameters of sugar cookies made with different concentrations of lentil flour was investigated, Table 9.

Table 9

Evolution of CIELab color parameters in sugar biscuits enriched with lentil flour during storage (results are presented as mean ± standard deviation)

Sample	Storage time, days	L*	a*	b*	ΔE*
CS	0	73.13±0.21 ^{i,j}	1.17±0.09 ^{c,d}	23.28±0.09 ^{b}	-
	10	73.75±0.11 ^{k,j}	1.36±0.07 ^{d,e}	24.17±0.04 ^{b}	1.01±0.01 ^{a}
	20	74.09±0.16 ^{k}	1.58±0.08 ^{e}	23.91±0.13 ^{b}	1.21±0.07 ^{a}
	35	75.04±0.17 ^{l}	1.37±0.08 ^{d,e}	22.48±0.10 ^{a}	2.08±0.05 ^{c}
SLF 1	0	72.44±0.11 ^{h}	1.48±0.07 ^{d,e}	25.33±0.04 ^{c}	-
	10	71.55±0.09 ^{g}	0.72±0.04 ^{b}	27.02±0.15 ^{d}	2.05±0.08 ^{b,c}
	20	73.55±0.13 ^{j}	0.22±0.02 ^{a}	23.01±0.11 ^{a}	2.86±0.11 ^{e}
	35	69.80±0.19 ^{c,d}	1.56±0.03 ^{e}	27.52±0.16 ^{d,e}	3.43±0.09 ^{g}
SLF 5	0	71.37±0.13 ^{f,g}	1.03±0.02 ^{c}	33.01±0.26 ^{j}	-
	10	72.38±0.10 ^{h}	0.98±0.01 ^{c}	32.71±0.18 ^{i,j}	1.05±0.05 ^{a}
	20	72.67±0.15 ^{h,i}	0.83±0.01 ^{b}	31.48±0.22 ^{h}	2.01±0.07 ^{b,c}
	35	73.01±0.13 ^{i}	0.71±0.04 ^{b}	30.37±0.19 ^{g}	3.12±0.09 ^{f}
SLF 10	0	70.62±0.14 ^{e}	1.53±0.05 ^{e}	29.51±0.15 ^{f,g}	-
	10	69.35±0.12 ^{c}	2.83±0.09 ^{g,h}	30.10±0.16 ^{g}	1.91±0.08 ^{c}
	20	69.88±0.20 ^{c,d}	2.10±0.01 ^{f}	27.40±0.07 ^{d,e}	2.31±0.06 ^{c,d}
	35	68.52±0.17 ^{a,b}	3.28±0.12 ^{i}	29.97±0.09 ^{g}	2.77±0.05 ^{e}
SLF 15	0	70.12±0.16 ^{d,e}	1.55±0.09 ^{e}	28.91±0.11 ^{g}	-
	10	69.75±0.15 ^{c,d}	2.63±0.05 ^{g}	29.10±0.08 ^{f}	1.16±0.03 ^{a}
	20	69.28±0.13 ^{c}	2.90±0.07 ^{g,h}	29.64±0.09 ^{f,g}	1.75±0.05 ^{b}
	35	68.72±0.10 ^{b}	3.30±0.05 ^{i}	29.97±0.13 ^{g}	2.48±0.02 ^{d}

Note: CS – control sample – 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour; L *—luminosity; a *—red/green component; b *—yellow/blue component; ΔE * - overall difference of color. Different letters ($^{a-l}$) designate statistically different results ($p \leqslant 0.05$).

It was found that the values of the luminosity L* in the CS were higher than in the SLF. Thus, on the next day after baking (1 day), the L* were as follows: CS (73.75), SLF 1 (72.44), SLF 5 (71.37), SLF 10 (70.62) and SLF 15 (70.12). Probably, the color of the lentil flour led to the decrease in luminosity of the sugar cookies. The values of the a* component were higher in the case of samples CLF, demonstrating the presence of red pigments, being in the range of 1.03-1.55. The highest values of a* were in samples SLF 10 and SLF 15, 1.55 and 1.53 respectively. In the case of the b* component, also the highest values were in the CLF samples due to the yellow pigments that ranged from 25.33 to 33.01. The storage time did not essentially influence the values of the color parameters for all the investigated samples.

The ΔE* values were analyzed to show whether there is a difference in the colors perceived by the human eye during the storage of the biscuits. According to Lo Faro et al. [91] , when keeping the CS for 20 days, the ΔE* values were in the range 1.01-1.21, which demonstrates a small difference in color (0.5 < ΔE* < 1.5), and in 35 days a difference color was barely distinguishable (2.08). In the case of the SLF1, during 20 days, the ΔE* values were in the range of 2.05-2.86, demonstrating a small difference in color (0.5 < ΔE* < 1.5), and in the 35th day, a color difference was very distinct (3.43), because it was within the limits 3 < ΔE* < 6. The SLF 5 sample kept for 10 days had the ΔE* values that demonstrated a small color difference (0.5 < ΔE* < 1.5), in the 20th day - a barely distinguishable difference (2 < DE < 3) and on the 35th day - a color difference was very distinct (3.12), as it was in the range 3 < ΔE* < 6. For the SLF10 and SLF 15 samples kept for 35 days, the ΔE* values changed from a small difference to barely distinguishable.

The microbiological analysis of the experimental samples of biscuits kept for 35 days are presented in Table 10.

Table 10

Influence of storage time on the microbiological indicators of sugar biscuits with added lentil flour

Microbiological indicators	**Storage time, days**	**Admitted level [66]**	**Experimental data**				
			CS	**SLF1**	**SLF5**	**SLF10**	**SLF15**
QMAFAnM, CFU, max.	1	1×10^4	$<1\times10^2$	$<2\times10^2$	$<2\times10^2$	$<1\times10^2$	$<1\times10^2$
	10		$<2\times10^2$	$<3\times10^2$	$<2\times10^2$	$<4\times10^2$	$<3\times10^2$
	20		$<5\times10^2$	$<1\times10^3$	$<7\times10^2$	$<6\times10^2$	$<5\times10^2$
	35		$<7\times10^3$	$<7\times10^3$	$<5\times10^3$	$<5\times10^3$	$<6\times10^3$
Coliform bacteria	1	Not allowed in 0.1 g	There were not found				
	10						
	20						
	35						
Fungi, CFU, max.	1	50	<1	<2	<2	<1	<1
	10		<5	<7	<5	<3	<3
	20		<10	<10	<15	<10	<10
	35		<40	<25	<20	<20	<25
Yeast, CFU, max.	1	50	<1	<2	<2	<3	<2
	10		<5	<5	<5	<5	<3
	20		<10	<10	<10	<10	<10
	35		<25	<20	<20	<20	<25

Note: QMAFAnM – quantity of mesophilic aerobic and facultative anaerobic microorganisms; CFU – colony-forming unit. CS – control sample – 100 % premium quality wheat flour; SLF1 – sugar biscuits with 1 % lentil flour; SLF5 – sugar biscuits with 5 % lentil flour; SLF 10– sugar biscuits with 10 % lentil flour; SLF 15– sugar biscuits with 15 % lentil flour; L *—luminosity; a *—red/green component; b *—yellow/blue component; ΔE * - overall difference of color.

The analysis of the total microorganisms number in the biscuit samples showed acceptable values throughout the storage period, which fall within the permissible limits. Based on these results, it can be said that the technological parameters used for baking were adequate, which influenced obtaining a microbiologically safe product for consumption.

4. Conclusions

To manufacture sugar biscuits enriched with RLF, both the sensory quality and composition of RLF were first evaluated. The effect of incorporating RLF (1–15%) on the kneading process, dough textural properties, as well as the quality, color parameters, and microbiological stability of biscuits during storage, was investigated. The results showed that increasing RLF content led to higher dough hardness, while cohesiveness and resistance decreased proportionally. Sensory analysis indicated that biscuits containing 1–10% RLF exhibited a firm texture, a pleasant and uniform appearance, and improved taste, aroma, and color. However, the addition of 15% RLF resulted in a pronounced lentil flavor. During storage, the water swelling capacity decreased by approximately 1.2 times. Microbiological analysis demonstrated no evidence of microbial growth or product deterioration throughout the storage period. Additionally, storage time had no significant effect on the color parameters of the analyzed samples. These findings suggest that the incorporation of RLF in sugar biscuit production is a viable approach to enhancing their nutritional value.

Acknowledgement. The authors would like to thank the Institutional Project 020405 “Optimizing food processing technologies in the context of the circular bioeconomy and climate change”, Bio-OpTehPAS, being implemented at the Technical University of Moldova.

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Citation: Boestean O., Bulgaru V., Netreba N., Ghendov-Mosanu A. (2026). Physicochemical, sensory and textural evaluation of sugar biscuits supplemented with lentil flour. *Journal of Engineering Science.* 2026, 33 (1), pp. 146-164. https://doi.org/10.52326/jes.utm.2026.33(1).10.

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