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Physical and Functional Characteristics of Extrudates Prepared from Quinoa Enriched with Goji Berry

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Abstract: In this research, the possibilities of applying the extrusion process to produce functional food from quinoa enriched with goji berries were studied. The physical (expansion ratio, bulk density, hardness, and specific mechanical energy) and functional (water solubility index, water absorption index) characteristics were determined and optimized by the response surface methodology. Extrudates were produced using a laboratory single-screw extruder. The full factorial experimental design was used ($N = 2^2$) with three complementary center points to show the interactions of the amount of goji berry (1, 3, and 5%) and the feed moisture content (13, 16, and 19%) of the mixture on the physical and functional characteristics. Increasing moisture content from 13 to 19% resulted in extrudates with a lower expansion ratio, water absorption index, water solubility index, specific mechanical energy, and higher density and hardness. Increasing the amount of goji berries from 1 to 5% led to a decrease in expansion ratio, water absorption index, and hardness, and an increase in density, water solubility index, and specific mechanical energy. Optimal extrusion conditions for production of extrudates from quinoa enriched with goji berry were 16.3% feed moisture content and 1.32% goji berry's amount.

Keywords: extrusion; quinoa; goji berry; physical characteristics; functional characteristics; optimization

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1. Introduction

Phenolic substances in plants possess various biological and nutritional effects [1,2]. Quinoa (*Chenopodium quinoa*) has a high protein content and a remarkable amino acid profile, for example, lysine and sulfur amino-acids; therefore, it has been assigned by NASA to the novel products with notable nutritional properties for long-duration missions in space [3]. The composition of quinoa also includes minerals, oils, starch, as well as several vitamins and antioxidants [4]. The authors of [5] confirmed the high antioxidant activity of the polyphenols in quinoa. The phenolic substances of quinoa were related to antioxidative, antiradical, and antitumor effects in the human body [6]. The availability of bioactive compounds in quinoa showed the possibility of its use for dietetic foods.

The goji berry (*Lycium barbarum* L.) traditionally originates from Asian countries where the fruits has been applied in alternative medicine and functional foods [7,8]. An ample amount of its composition consists of polysaccharides, and their presence is related to various biological properties, for example, rejuvenating and reducing the effects of fatigue, neuroprotective effect, improved metabolism, glucose reduction for diabetics, antioxidant and anti-tumor ability, increased immunity, etc. [8,9]. It was established that the goji berries improved the feelings of overall well-being, tiredness, stress, neurological, stomach and muscular problems, blood pressure, visual capacity [10,11], plasma values of anti-oxidants [12], and immune factors [13].

The extrusion process has been widely used all over the world for the production of expanded foods, breakfast cereals, and pet foods [14]. This process possesses many advantages in comparison to other food technologies due to the short processing time and its continuous nature [15]. Despite the popularity of the extrusion processes in food and feed production, there are still missing data concerning the raw materials and extruder conditions of some products [16]. The investigations of the authors of [15,17] showed that quinoa can be successfully used for production of extruded foods. The authors of [15] extruded quinoa flour and investigated the effect of feed moisture content, temperature, and screw speed on the physicochemical properties of extrudates. Similar investigations were carried out in [17] for ten novel quinoa breeding lines. The incorporation of fruits in extrudates improves not only the content of healthy compounds but also the attractive appearance of the final product [18]. The authors of [19] used different fruits in extruded snacks for children's diets and studied the interactions between the type of fruit (banana, apple, strawberry, tangerine), extrusion process, and the physical properties of the resulting product. The incorporation of the goji berry in extrudates results in products with a high content of bioactive substances and high antioxidant activities [18,20]. The authors of [18] added dried goji berries in a mixture of rice flour, wheat flour, sucrose, and whey protein and investigated the physical properties and antioxidant activity of the extruded product. Similar investigations were carried out in [20], where various amounts of goji berry powders (from 3 to 20%) were added to rice flour. According to [21], the carbohydrates, proteins, and fibers in the raw material influenced the properties of the extruded products. The same authors enriched corn grits with dried goji berries and cranberries (from 1 to 5%) and studied the functional characteristics of the extrudates—the water absorption index (WAI), water solubility index (WSI), and color characteristics. There is no found literature on the enrichment of quinoa extrudates with goji berries.

The main objectives of this investigation were: (1) to determine the effect of extrusion variables such as the amount of goji berries in raw material and feed moisture on the physical and functional characteristics of an extrudate; (2) to optimize the amount of goji berry and feed moisture content in order to obtain a product ready for incorporation in new healthy foods with minimum production costs.

2. Materials and Methods

2.1. Materials

The study was conducted with quinoa from the local market and goji berries produced in a private Bulgarian farm. The quinoa seeds were milled using a laboratory stone mill (Predom, BG Agro Ltd., Letnitsa, Bulgaria), up to an average particle size of 0.245 mm. The goji berries were dried in a convective oven at 100 °C up to a water content of 15% w.b. ± 0.25% w.b. [22]. After drying, the fruit was milled using a laboratory mill and then incorporated and homogenized manually in the quinoa as a percentage ratio of 1, 3, and 5%.

The initial moisture content of the mixture from quinoa and goji berry was determined by drying for 24 h at a temperature of 105 °C in a drying oven Labster 136N250 (Barcelona, Spain) [23]. The desired moisture contents were achieved by adding distilled water to the initial mixture. The quantity of added water was calculated by the following formula:

$$m_w = \frac{w_d - w_{in}}{100 - w_d} * 100 \quad (1)$$

where m_w is the quantity of added water, g water/100 g product; w_d is the desired moisture content, % w.b.; and w_{in} = 12 ± 0.10% w.b is the initial moisture content of the mixture.

The mixtures were stirred for 10 min and stored in polyethylene bags for 12 h to equalize the moisture at refrigerating conditions of 5 °C. The samples were homogenized manually again before extrusion to avoid cohesion between particles.

2.2. Methods

2.2.1. Regression Modeling and Statistical Analysis

A response surface methodology was applied using a full factorial experimental design (N = 2^2) with three center points, in 7 runs. Table 1 shows the independent variables (amount of goji berry and feed moisture content), and their levels were chosen using preliminary experiments and literature data. The dependent response variables were: expansion ratio (ER), bulk density (BD), water absorption index (WAI), water solubility index (WSI), hardness (HD), and specific mechanical energy (SME). They were determined on the basis of three replications.

Table 1. Experimental design.

Number	Natural Levels		Coded Levels	
	Amount of Goji Berry, %	**Moisture Content, % w.b.**	X_1	X_2
1	1	13	−1	−1
2	5	13	+1	−1
3	1	19	−1	+1
4	5	19	+1	+1
5	3	16	0	0
6	3	16	0	0
7	3	16	0	0

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Regression models for the dependent variables were obtained using the StatGraph v2.0 statistical software at a level of significance of 0.05:

$$Y = b_0 + b_1 X_1 + b_2 X_2 + b_{12} X_1 X_2 \quad (2)$$

where Y is the experimental response, b_0 is the coefficient for intercept, b_1 and b_2 are linear coefficients, b_{12} is the interactive coefficient, and X_1 and X_2 are independent variables.

For the comparison of the experimental runs, Fisher's least significant difference test at a significance level of 0.05 was used in Excel 2010 with a one-way analysis of variance (one-way ANOVA).

2.2.2. Extrusion

A single-screw laboratory extruder Brabender 20 DN (Duisburg, Germany) was used to carry out the extrusion at the following conditions: die diameter 3 mm; feeding screw speed 30 min^{-1}; extruder screw speed 200 min^{-1}; compression ratio 3:1; and temperatures in the three zones of the extruder were 150, 160, and 170 °C, respectively. After extrusion, the extrudates were cooled to room temperature (20 ± 1 °C) and placed in polyethylene bags until measurements of the physical and functional characteristics.

2.2.3. Determination of Physical Characteristics

The ER was determined as a ratio of the diameter of the extrudate measured with digital vernier caliper and the die diameter (3 mm):

$$\text{ER} = \frac{diameter\ of\ extrudate}{diameter\ of\ extruder\ die} \quad (3)$$

For determination of the BD, assuming a cylindrical shape of extrudate, the diameter and length of the sample were measured using a digital vernier caliper, and the weight of the sample using an analytical balance (Kern ABS 220-4N with level of accuracy 0.1 mg). Then, the BD was calculated using the following equation:

$$\text{BD} = \frac{weight\ of\ extrudate}{volume\ of\ extrudate} \tag{4}$$

The HD (force) of the extrudates was determined using a TA-XT texture analyzer (Stable Micro Systems Ltd., Godalming, UK) equipped with 50 kg load cell and 2-bladed Kramar shear cell (distance 10 mm, speed 1.0 mm/s). The hardness was determined as the minimal force (N) necessary to crumb the sample.

For determination of SME, the torque and the screw speed were adjusted and measured with a DCE 330 device, a part of the extruder equipment. The mass flow rate was determined at each replication by measuring the weight of the obtained extrudate for 10 s. The SME (Wh/kg) was determined by the equation:

$$\text{SME} = \frac{M.n.\pi}{30.Q} \tag{5}$$

where M is torque, N.m; n is speed of the extruder screw, min^{-1}; and Q is mass flow rate, kg/h.

2.2.4. Determination of Functional Characteristics

The WSI (%) and WAI (g/g product) were determined using the procedure described in [24] with modifications: the ground extrudate was placed in falcon tubes with a constant weight and then dispersed (0.2 g) in 5 mL of distilled water. The sample was placed in water bath at 30 °C for 30 min, with gentle stirring during this period, followed by centrifugation at 3000 min^{-1} for 20 min using a centrifuge CH 90-2A (Polypharma Sarl, Douala, Cameroun). The supernatant was moved to an evaporating dish and then dried at 105 °C in a drying oven until a constant weight was achieved.

The WSI was calculated as follows:

$$\text{WSI} = \frac{m_{ds}}{m_s}100 \tag{6}$$

The WAI was calculated as follows:

$$\text{WAI} = \frac{m_g}{m_s} \tag{7}$$

where m_{ds} is weight of dry solids after evaporation of the supernatant, g; m_s is weight of sample, g; and m_g is weight of sediment, g.

2.2.5. Optimization of Physical and Functional Characteristics

The aim of the optimization was to improve the physical and functional characteristics of the extrudates from quinoa enriched with goji berries in order to obtain a well-puffed extrudate suitable for long-term storage and produced with minimum costs (energy). A numerical multiple response optimization technique was used and applied [24] to determine the optimal combination of the goji berries' amount and the feed moisture content. The assumptions were to create a product with maximum ER and minimum BD, WAI, WSI, HD, and SME.

3. Results and Discussion

3.1. Experimental Results

Table 2 shows the mean values, standard deviations, and statistically significant equalities/differences (between the experimental runs) of the physical and functional characteristics. The results showed that the ER varied between 1.93 and 2.15, the BD between 0.146 and 0.268 g/cm^3, the WAI between 6.05 and 7.13 g/g, the WSI between

17.13 and 23.03%, the HD between 2.58 and 3.47 N, and the SME between 36.19 and 81.12 (Wh)/kg.

Table 2. Experimental results for physical and functional characteristics of extrudates.

№	ER	BD, g/cm^3	WAI, g/g	WSI, %	HD, N	SME, Wh/kg
1	2.15 ± 0.05 ^{a}	0.146 ± 0.004 ^{a}	8.05 ± 0.06 ^{a}	21.78 ± 0.27 ^{a}	24.21 ± 0.94 ^{a}	71.08 ± 1.99 ^{a}
2	1.93 ± 0.06 ^{b}	0.170 ± 0.004 ^{b}	7.51 ± 0.12 ^{b}	25.61 ± 0.53 ^{b}	17.24 ± 1.33 ^{b}	81.12 ± 2.01 ^{b}
3	1.93 ± 0.03 ^{b}	0.195 ± 0.004 ^{c}	6.53 ± 0.14 ^{c}	20.68 ± 0.39 ^{c}	29.93 ± 0.32 ^{c}	36.19 ± 1.36 ^{c}
4	1.93 ± 0.01 ^{b}	0.268 ± 0.012 ^{d}	6.34 ± 0.12 ^{c}	19.46 ± 0.22 ^{d}	23.09 ± 1.02 ^{a}	55.44 ± 2.22 ^{d}
5	2.02 ± 0.02 ^{c}	0.205 ± 0.010 ^{c}	7.07 ± 0.03 ^{d}	22.42 ± 0.26 ^{e}	19.60 ± 0.51 ^{d}	54.67 ± 2.28 ^{d}
6	2.08 ± 0.05 ^{ac}	0.156 ± 0.006 ^{a}	7.07 ± 0.01 ^{d}	21.98 ± 0.13 ^{ae}	21.44 ± 0.51 ^{e}	54.32 ± 2.34 ^{d}
7	1.94 ± 0.05 ^{b}	0.203 ± 0.006 ^{c}	7.02 ± 0.02 ^{d}	21.97 ± 0.07 ^{a}	18.20 ± 0.65 ^{b}	58.21 ± 1.16 ^{d}

Different lowercase letters (a, b, c, d, e) show a significant difference between the experimental runs ($p < 0.05$).

The standardized Pareto charts with the effects of each independent variable for the physical and functional characteristics are presented in Figure 1. Feed moisture was the main factor that affected all investigated characteristics except for the HD. This confirms the results in [25–27]. The interactive coefficient was insignificant for BD and HD at a level of significance of 0.05. For ER, the interactive coefficient was equal as an absolute value to the others. For WSI and SME, the interactive coefficient was higher than coefficient b_1 in Equation (2).

3.2. Expansion Ratio

ER and BD are determined to directly classify the expanded extrudate characteristics. Extruded products desirable to consumers have a well puffing. The most desired characteristics for expanded products are high expansion and low density [15]. The quinoa has lower expansion ratio values [15,17] compared to other materials, which may be explained by fact that the quinoa flour had higher amounts of protein, fiber, and lipids in comparison to other cereal grains. The Pareto charts (Figure 1) showed that all effects were significant for the expansion ratio, and both investigated variables influenced approximately equally and negatively on the expansion. The decrease in the moisture resulted in a decrease in the degree of starch gelatinization, which limited the expansion [27]. Under the extrusion of the raw material with higher moisture, it could not evaporate sufficient water, which resulted in products with a higher BD [28]. The authors of [20] established that the addition of 3 and 7% of goji berries in rice semolina led to an increase in the ER, whereas higher addition levels led to a significant decrease in the ER. In our study, a similar maximum was not observed. The effect of two investigated variables on the expansion ratio of extrudates is shown in Figure 2. The figure shows that the expansion decreased by approximately 10% at a low amount of goji berries with an increase in the moisture content. At the high level of the goji berries' amount, the expansion ratio remained constant with the increase in the moisture content. The effect of the goji berries' amount was clearly expressed at a low level of moisture content and was approximately imperceptible at a high level of moisture. The expansion depended on the feed moisture, and this would affect the viscoelastic characteristics of the starch-based material. The level of feed moisture content during extrusion provoked changes in the amylopectin molecular structure of the material, reducing the melt elasticity and decreasing the expansion of the extruded product [27]. The authors of [29] reported that low feed moisture content may limit the flow of the material and increase the shearing rate and residence time, leading to an increase in the level of gelatinization and expansion.

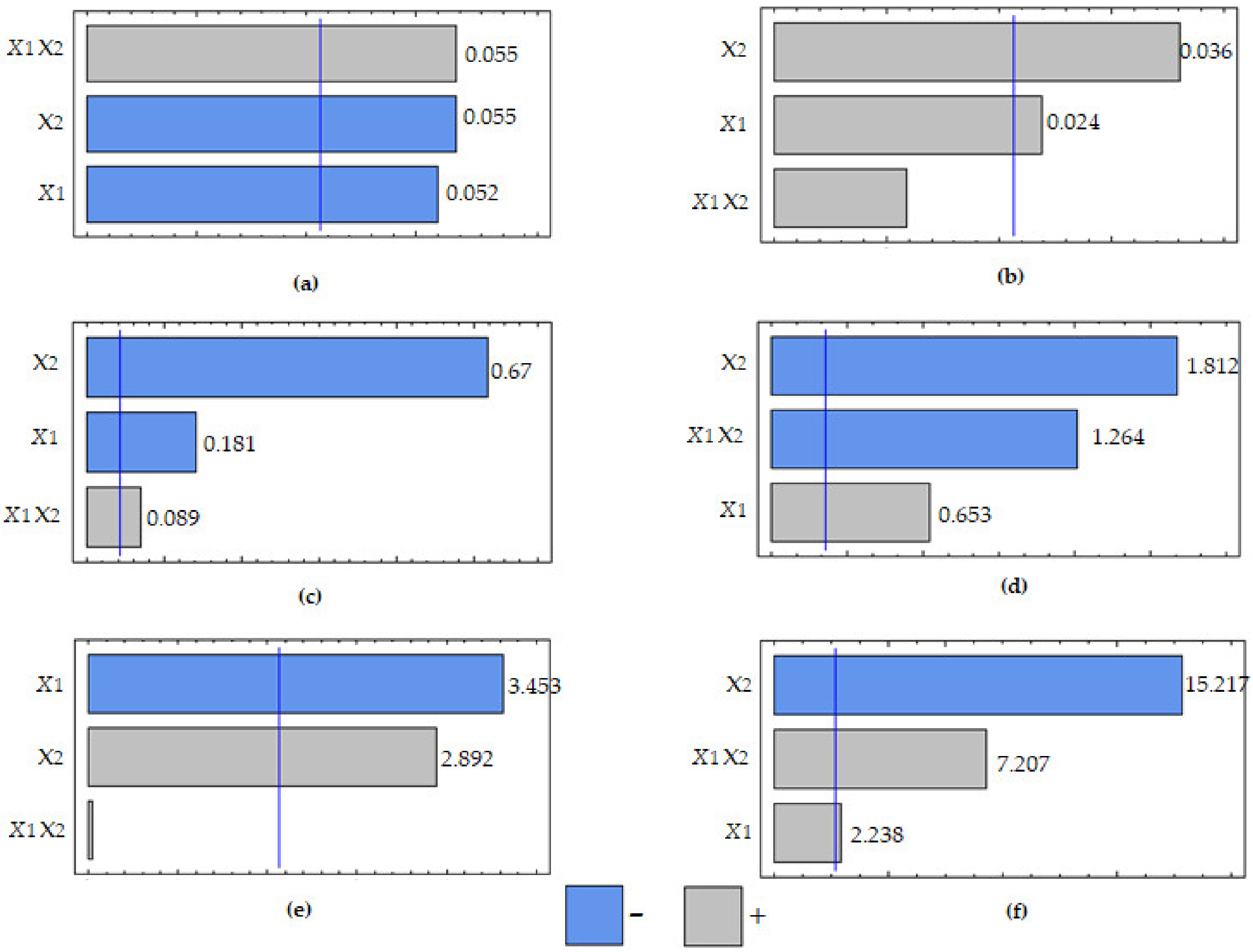

Figure 1. Standardized Pareto charts for the effects of the amount of goji berry (X_1) and feed moisture content (X_2) on the physical and functional characteristics ((**a**): ER; (**b**): B; (**c**): WAI; (**d**): WSI; (**e**): HD; (**f**): SME). The vertical line in the chart tests the significance of the effects at a level of significance = 0.05.

3.3. Bulk Density

The dependence of the density on the two independent variables is presented in Figure 2. The highest value of the BD was observed at high levels of goji berry and moisture. The extruded products with lower expansion possessed higher BD [30]. The lowest BD was observed at low levels of these two independent variables. The moisture of the raw material had a greater impact on the BD in comparison to the amount of goji berry. It was approximately 1.5 times more pronounced, compared to the goji berry. Density is an indicator that shows what kind of expansion has occurred as a result of extrusion. There is an inverse relationship between the expansion and BD—extrudates with a smaller degree of expansion have a higher BD [31]. According to the authors of [27], the increase in the feed moisture content can lead to a modification of the amylopectin and amylose structure of the material, which reduces elasticity characteristics of the dough through plasticization of the melt, resulting in reduced gelatinization. This leads to a decrease in the expansion and an increase in the density of extrudates. The formation of a complex between the amylose and native lipids presented in cereal starches may limit the swelling, leading to a more rigid structure and an increase in the density [32]. The increase in the BD with the increase in goji berry could be explained with higher levels of proteins and lipids in extrudates. The authors of [19] investigated the physical properties of extrudates enriched with different fruit powders. They estimated that the rise in the BD of extrudates was attributed to the availability of sugar and soluble fiber absorbing more moisture, thus leading to a decrease in the expansion.

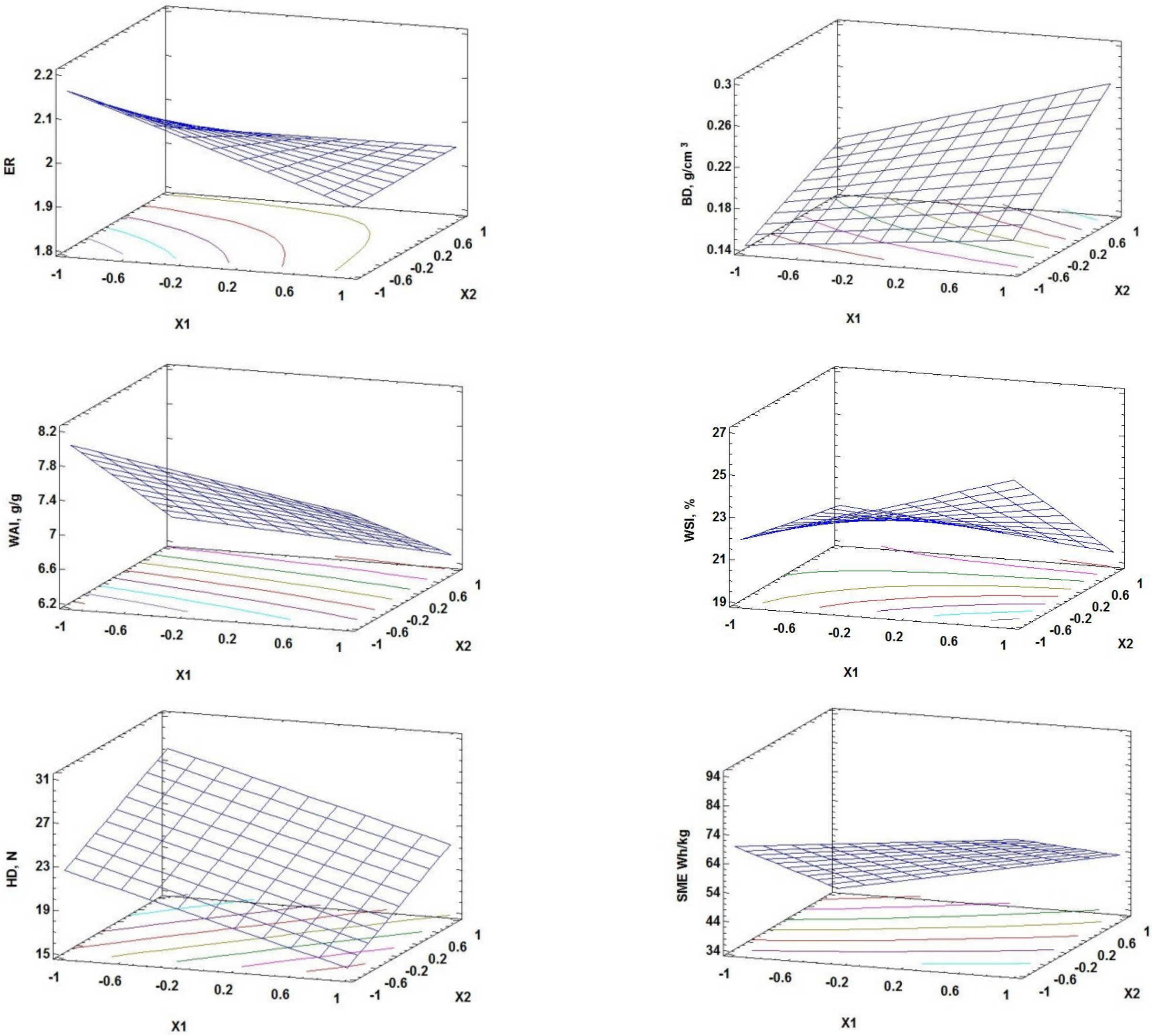

Figure 2. Response surfaces for the physical and functional characteristics (X_1: amount of goji berry; X_2: feed moisture content).

3.4. Water Absorption Index

The WAI characterizes the quantity of water absorbed by starch and can be applied for the determination of the level of gelatinization because native starch is insoluble in water at room temperature [27,33]. The two independent variables influenced negatively on the WAI, as the effect of the moisture was more pronounced. When the two variables increased simultaneously, WAI decreased from 8 to 6.3 g/g product (Figure 2). Higher moisture content likely leads to an increase in starch dextrinization, and this limits its gelatinization and leads to a decrease in WAI. The increase in the goji berries' quantity also contributes to this process due to the increase in lipids in the semolina that interacts with the starch and limits its gelatinization [34]. The authors of [15] established values of WAI from 2.33 to 3.05 g/g for extrudates from the quinoa var. Cherry Vanilla. The difference with our results may be due to use of a twin-screw extruder as well as the composition of the quinoa. The authors of [21] found a positive relationship between the amount of goji in corn grits and WAI. In contrast to those results, and in accordance with our results, the authors of [35] found significantly lower WAI in extrudates from rice and corn when the amount of the açaí berry increased. They attributed this to the reduction in starch content, causing limited gelatinization during extrusion process of the samples with the addition of the açaí fruit. Another explanation could be given with the increase in protein content by the increasing fruit proportion causing bonds with amylose and amylopectin, thus decreasing the ability of starch to absorb water molecules.

3.5. Water Solubility Index

The WSI is related to the destruction of molecular bonds and an indicator for the quantity of soluble components after extrusion [29]. At low level of goji berries, the effect of moisture content on the WSI was slightly expressed. When the moisture of the semolina increased, the WSI decreased considerably at a high level of goji berries. This is in accordance with the results in [15], where the authors found the same trend during the extrusion of the quinoa var. Cherry Vanilla. The quantity of goji berry in the semolina influenced the WSI positively, but the coefficient in Equation (2) in Figure 1c is smaller than this moisture, which leads to a negative combined effect of these two variables. This conclusion is confirmed by the response surface (Figure 2). The authors of [21] established similar values of WSI of extrudates from corn grits and goji berries, but, in contrast to our investigation, they found no significant effect from the amount of goji berries on the WSI in the range of the experiment. At a low level of moisture, with the increase in goji berries, WSI increased, while WSI decreased at high level of moisture, i.e., the effect of moisture prevailed. A possible reason for the divergent influence of goji berries at different moistures is the significant interfactorial interactivity. The rise in WSI with the increase in the amount of goji berry could be explained by the presence of soluble carbohydrates and dietary fibers in goji berries, established in [36].

3.6. Hardness

The HD of extrudates, expressed by the minimal force necessary to break the sample, is very important for the product from a technological point of view. It is established that the feed moisture content has a notable effect on the hardness and crispiness of the final product [27,37,38]. Moisture is the variable that thoroughly influences the expansion and BD of the extrudates. It was observed that a product with higher BD and lower expansion is harder [33]. The initial moisture of the material is important for the generation of vapor bubbles in the melted mass when it exits from the extruder die. The pressure in the extruder die decreases with the rise in the moisture content, which contributes to denser and harder product [38,39]. The response surface (Figure 2) shows that the highest level of hardness was obtained at a low level of goji berries and a high level of moisture. The authors of [40] found a decrease in the hardness of extrudates produced from the blend of barley and tomato pomace at a die temperature of 140 °C when the amount of the tomato pomace increased. However, at a higher die temperature, the increase in the amount of tomato pomace led to an increase in the hardness. Our results showed that the lowest hardness was obtained at a high level of goji berry and low moisture levels. The HD increased when the moisture increased, while the increase in goji berry led to its decrease. This conclusion is confirmed by the coefficients in Equation (2) and Figure 1e—negative for goji berry and positive for moisture. The negative effects of the goji berries could be explained by the gelling effect of the carbohydrates presented in goji berries. There are investigations, such as [19], where the authors found a directly proportional relationship between the hardness and the fruit amount, but there are no investigations concerning the hardness of extrudates from quinoa with goji berries.

3.7. Specific Mechanical Energy

The size of consuming energy influences the degree of macromolecular transformations and interactions taking place during extrusion, namely, the degree of gelatinization of starch and the rheological properties of the melted material [38]. On the other hand, the high SME is related to higher production costs. The effects of the two independent variables on SME are shown in Figure 2. The consumed specific mechanical energy was lower at a low amount of goji berries and high amount of moisture. The higher moisture content led to less pronounced shear stresses in the cylinder of the extruder. Consequently, the torque decreased. This facilitated the movement of the dough inside the extruder and increased the mass flow rate. The homogeneity of the mixture turned out to be an important condition during extrusion. In [19], it was established that the addition of different fruit powders reduced the values of the SME. The authors explained this with the lubrication effect of sugars and fibers in the fruits. On the other hand, the authors of [41] found insignificant effects from the amount of tomato pomace on the SME. In our study, the Pareto chart (Figure 1f) showed a slightly expressed positive effect of the amount of goji berries on the SME. The probable reason is that the addition of goji berries increased the viscosity of the dough during extrusion. It also increased the residence time of the product in the extruder, which had a negative effect on the mass flow rate. This also led to increased gelatinization of the product. The combination of a lower moisture content and a high concentration of goji berries increased the SME significantly.

3.8. Optimization

The results from the response optimization showed that the goji berries of 1.32% and the feed moisture content of 16.3% was the optimal combination of independent variables and could be used for the production of quality extrudates. The optimization showed that it is possible to add a small amount of dried goji berry without a significant deterioration of the physical and functional characteristics of the extrudate. The values of the response variables at the optimal combination of the amount of goji berries and feed moisture content are shown in Table 3.

Table 3. Physical characteristics of extrudates at the optimal combination of independent variables.

Response Variable	Value
ER	2.03
BD	0.17 g/cm^3
WAI	7.16 g/g
WSI	21.36%
HD	25.19 N
SME	51.28 Wh/kg

4. Conclusions

The physical and functional characteristics of extrudates from quinoa enriched with goji berries depended on the goji berries' amount and the feed moisture content. The increase in moisture content from 13 to 19% resulted in extrudates with a lower expansion ratio, water absorption index, water solubility index, specific mechanical energy, and a higher density and hardness of the extrudates. The increase in goji berry's amount from 1 to 5% caused a decrease in the expansion ratio, water absorption index and hardness, and an increase in density, water solubility index, and specific mechanical energy. Optimal extrusion conditions to produce extrudates from quinoa enriched with goji berry were established at 16.3% feed moisture content and 1.32% goji berry amount. The results showed that it is possible to add a small amount of dried goji berry without a significant deterioration of the physical and functional characteristics of the extrudate.

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Abbreviations

ER	Expansion Ratio
BD	Bulk Density
WAI	Water Absorption Index
WSI	Water Solubility Index
HD	Hardness
SME	Specific Mechanical Energy

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