#### **ACCEPTED ARTICLE** 1 Production of functional pasta with low phenylalanine based on potato and 2 tapioca starches 3 4 5 Short Title: Pasta for phenylketonuria patients 6 Alireza Rezaei<sup>a</sup>, Mania Salehifar<sup>a\*</sup>, and Vajiheh Fadaei Noghani<sup>a</sup> 7 8 <sup>a</sup> Department of Food Science and Technology, Shahr-e-Qods Branch, Islamic Azad University, Tehran, Islamic Republic of Iran. 9

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# 12 ABSTRACT

13 Phenylketonuria (PKU) is a genetic disorder necessitating a low-protein and phenylalanine diet. 14 This study aimed to explore the feasibility of producing a low-protein pasta using potato and 15 tapioca starches. The pasta formulation substituted semolina flour with a blend of potato and 16 tapioca starches. Date kernel fiber and xanthan gum were incorporated as prebiotic compounds 17 and texture enhancers, respectively. Physicochemical (moisture, fat, total ash, protein, 18 phenylalanine, cooking loss, cooking time, color indexes, and hardness) and sensory properties 19 (texture, flavor, color, and overall acceptability) were evaluated and compared against the control 20 sample (based on semolina flour). The results demonstrated no significant alteration in moisture 21 and fat content upon substitution, but a significant decrease in ash and protein content (p < 0.05). 22 Consequently, phenylalanine levels decreased from 530.58 mg/100 g in the control sample to 23 24.49-26.60 mg/100 g in the pasta. Replacing flour with starches increased cooking loss, reduced 24 cooking time, and diminished pasta hardness compared to the control (p<0.05). The pasta exhibited higher L\* and lower a\* and b\* values than the control. Sensory evaluation revealed that the pasta 25 26 containing 35% potato starch and 40% tapioca starch attained the highest scores, indicating its 27 favorable acceptability. Overall, this study suggests that the combination of potato and tapioca 28 starches, along with date kernel fiber and xanthan gum, enables the production of and low-protein 29 pasta suitable for PKU patients.

30 Keywords: Date kernel fiber, Low-protein pasta, Phenylketonuria, Potato starch, Tapioca starch.

# 31 **1. INTRODUCTION**

Pasta is one of the most popular and common staple foods in the human diet, with convenientconsumption, favorable organoleptic characteristics and high nutritional value. Pasta contains

34 digestible carbohydrate and has little fat. In addition, pasta has low price and its shelf life is long.

35 The main ingredients of pasta formulation are durum wheat flour (semolina) and water (Bresciani,

36 Pagani et al. 2022).

Phenylketonuria (PKU) is a genetic disorder caused by the deficiency of the phenylalanine hydroxylase enzyme. This enzyme causes the conservation of phenylalanine to tyrosine, and the lack of this enzyme causes an increase in the accumulation of phenylalanine in the blood and brain (MacDonald, Van Wegberg et al. 2020). PKU can lead to sever neurological problems such as irreversible mental disability if not controlled. The complications of this disease can be prevented by severely limiting protein-containing foods and consuming food products with low phenylalanine content (McWhorter, Ndugga-Kabuye et al. 2022).

44 Pasta cooking quality is influenced by protein content, with gluten being crucial for elasticity and chewability. Gluten-free pastas can be made using an appropriate ratio of proteins, hydrocolloids, 45 46 and water. Research focuses on developing gluten-free products using non-wheat flours, dairy products, and emulsifiers to improve structure, mouth feel, acceptability, and shelf-life. However, 47 48 issues include the absence of gluten and achieving acceptable quality without traditional wheat 49 ingredients (Ungureanu-Iuga, Dimian et al. 2020, Gasparre and Rosell 2023). To develop gluten-50 free pasta, polymer ingredients such as starches and hydrocolloids are needed to imitate the 51 viscoelastic behavior of gluten in the product batter and create a favorable texture, mouth feel and 52 acceptability (Zoghi, Mirmahdi et al. 2021, Zhang, Zhang et al. 2022). Xanthan gum is one of the 53 most widely used hydrocolloids for the development of gluten-free or gluten-reduced products 54 (Palavecino, Bustos et al. 2017). Potato (Solanum tuberosum L.) is an excellent source of 55 carbohydrates and its starch content is about 75-80% based on dry weight (Dupuis and Liu 2019). The unique features of potato starch compared to cereal starches include the longer chain length 56 57 of amylopectin and amylose, greater granular size and purity, ability to exchange specific cations 58 with an effect on viscosity behavior, presence of phosphate ester groups on amylopectin, and the 59 formation of a strong viscoelastic gel. These unique features determine the sensory attributes and quality of gluten-free pastas (Raj, Dalal et al. 2020). Tapioca or cassava starch is a gluten-free 60 61 product that has favorable characteristics for application in the food industry. This starch has a 62 high availability and low price, and it controls or changes the characteristics of foods such as 63 appearance, texture, consistency, and storage stability (Reddy, Kimi et al. 2016).

64 The substitution of wheat flour with starch in gluten-free products can reduce fiber and nutrient 65 intake. To enhance the nutritional value of these products, various sources of dietary fiber and 66 nutritional compounds are used, with fruits and vegetables being crucial sources of dietary fiber (Zhang, Dongye et al. 2023). Dietary fibers are among the prebiotic compounds, and are 67 68 indigestible and have various health benefits, especially the regulation of human intestinal activity. 69 So, recently attention has been directed towards the development of prebiotic products (Abedinia, 70 Alimohammadi et al. 2021). Date fruit belongs to Arecacea family, and is native to tropical and 71 subtropical regions (Hussain, Farooq et al. 2020). Date consist of pulp and kernel, and the kernels 72 make up 5% to 15% of the total weight of fruit. Date kernel is the most important waste of date 73 processing factories, and is rich in fat, protein and dietary fiber (Alharbi, Raman et al. 2021). The 74 functional characteristics of date kernels have been found to consist of 22.5-80.2% dietary fibre, 75 3.1-7.1% moisture, 2.3-6.4% protein, 5.0-13.2 fat, and 0.9-1.8% ash. Due to their high dietary 76 fibre content, date seeds have a significant nutritional value and may be used to manufacture dishes 77 high in fibre and as dietary supplements. Date seeds have a high dietary fibre content since they 78 are generated in huge quantities as a waste product (Samea and Zidan 2019).

In this research, for the production of functional pasta with reduced protein and phenylalanine, wheat flour was replaced with different levels of potato (PS) and tapioca starches (TS), and xanthan gum and date kernel fiber (DKF) were used as texture improver and prebiotic agent, respectively, and the effect of starches on the physicochemical properties, cooking quality, sensory characteristics and protein and phenylalanine content of the produced pastas were evaluated.

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# 2. MATERIALS AND METHODS

### 86 2.1. MATERIALS

Semolina flour was prepared from Zar Flour of Karaj (Iran). Potato and tapioca starches were also purchased from Stage Company (Iran). Xanthan gum,  $\beta$ -carotene and date kernel fiber were purchased from Provisco Company (Germany), Merck Company (Germany) and Flavinea Company (Iran), respectively. The chemical composition of semolina flour, PS, TS and DKF used in this research was determined using the AACC standard method (AACC 2000) and their values are listed in Table 2. All the chemicals used for the tests were also purchased from Merck (Germany).

### 96 **2.2. Preparation of pasta treatments**

97 The formulation of the control pasta (penne) was as follows: 76% semolina flour, 1% DKF, 98 0.015% β-carotene, and 23.985% water. In the formulation of low-protein pastas, the combination 99 of different levels of PS and TS (Table 1) completely replaced semolina flour. Xanthan gum (1%) 100 was used as a texture improver. To prepare pasta treatments, dry ingredients were first mixed 101 together in a mixer (Anselmo, Italy) for 3 min.  $\beta$ -carotene was dissolved in the formulation water 102 and gradually added to the dry ingredients and stirred for 20 min. The resulting mixture was 103 extruded (La Monferrina Model P6, Roma, Italy) has an 8 mm x 3.4 mm cylinder-shaped diameter 104 at 45°C and under pressure of 0.6 mm Hg. During the extrusion process, the temperature of the 105 batter taken out of the mold was subjected to a flow of water at 20°C so that the pasta doesn't stick 106 together and doesn't losses its shape. The produced pastas were collected in a basket and placed 107 in a dryer for 24 h at a variable range of 20-80°C until they were completely dry and their moisture 108 content was less than 12%. After cooling, the produced pastas were packed in polypropylene bags 109 and kept at 18°C. Figure 1 shows the samples prepared for this study.

- 110 **Table 1.** The formulations of pasta treatments (g/100 g dry basis) with constant amounts of  $\beta$ -
- 111 Carotene, DKF, semolina flour (0.015, 1, and 75%, respectively) and water (in the amount to form

112	a dough).

Treatments	PS	TS	Xanthan gum
T0 (control)	-	-	-
T1	75	-	1
T2	65	10	1
T3	55	20	1
T4	45	30	1
T5	35	40	1
T6	25	50	1
T7	15	60	1
Т8	-	75	1

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DKF: date kernel fiber, PS: potato starch, TS: tapioca starch.

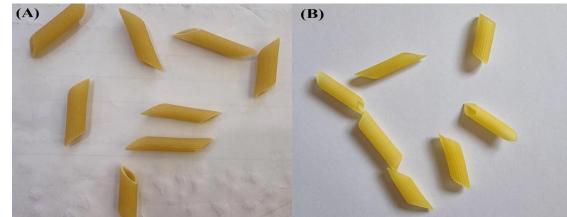


Figure 1. Pasta treatments (A) control; (B) pasta containing 35% PS+40% TS.

### 116 **2.3.** Analysis of physicochemical properties of pasta

117 2.3.1. Proximate and physicochemical analysis

118 Moisture, fat, total ash and protein content of pastas were determined using the AOAC standard 119 method (44-16, 30-01, 44-15, and 12-46, respectively) (AACC 2000). The cooking loss values of 120 the pasta samples were measured using the Iranian national standard method number 213 by 121 dividing pastas weight after by before of cooking. The color analysis of the raw pastas was done 122 through three indexes of L\* (bright), a\* (red/green) and b\* (yellow/blue) and using a colorimeter 123 (Hunterlab, America) (Milde, Chigal et al. 2020). The hardness of the cooked pastas was measured 124 using a Brookfield CT3 texture analyzer (England) with a cylindrical probe (35 mm) and the test 125 of single compression. Compression up to 80% of the initial thickness of the pasta was done by 126 the device probe at a speed of 1 mm/s, and the force required for this work was reported as N 127 (Piwińska, Wyrwisz et al. 2016).

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## 129 2.3.2. Measurement of phenylalanine

130 To measure the phenylalanine content of raw pastas, an HPLC device (Waters, America) 131 equipped with a UV detector and RP-C18 column with particle size of 5 µm, and dimensions of 132  $150 \text{ mm} \times 39 \text{ mm}$  was used. Initially, the sample (0.5 g) was mixed with 6 M HCl (8 mL) and after 133 airing, the resulting mixture was hydrolyzed in an oven for 22 h at a temperature of 110 °C. The 134 obtained hydrolyzates were first centrifuged at 5000 rpm for 10 min, neutralized by NaOH 3M 135 and then injected into HPLC device in a volume of 20  $\mu$ L. The mobile phase consisted of a mixture 136 of acetonitrile and phosphate buffer (pH = 3.5) in a ratio of 98 to 2, and its flow rate was set at 0.8 137 mL/min (Yaseen and Shouk 2011).

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# 2.4. Sensory evaluation of pasta

The sensory evaluation of the cooked pasta was done using 10 trained panelists and according to the 7-point Hedonic test (7= excellent sample and 1=very bad sample), and the panelists rated the texture, flavor, color and overall acceptability of the pasta treatments. Preparation of pastas for sensory evaluation was done by heating sample (50 g) in boiling water (250 mL) with 2% salt according to Table 4, for each sample separately and was evaluated by the panelists (Shogren, Hareland et al. 2006).

### 148 2.5. Statistical analysis of data

The results of the experiments were analyzed to investigate the significant difference between the data through one-way analysis of variance using SPSS 22.0 software. Duncan's multi-range test was used to compare the mean of the treatments at the 95% probability level.

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## 153 **3. RESULTS AND DISCUSSION**

### 154 **3.1. Chemical composition of pasta**

155 The results of examining the chemical composition of pasta (Table 3) showed that the amounts 156 of moisture, ash, fat and protein of pasta treatments were in the range of 10.31-10.83%, 0.61-157 0.72%, 1.10-1.14%, and 0.94-11.13%, respectively. Substitution of semolina flour in pasta 158 formulation with the combination of starches, DKF and xanthan gum didn't have a significant 159 effect on the moisture and fat content of produced pastas, but significantly reduced the amounts of 160 ash and protein (p<0.05). The decrease in the ash content of pasta is probably because the amount 161 of total ash in PS (0.23%) and TS (0.12%) is less than that of semolina flour (0.74%). The lower 162 protein content of PS (0.17%), TS (0.14%) and DKF (2.23%) compared to semolina flour (13.62%) 163 is also the reason for reducing the protein content of pasta with flour replacement. Yaseen and 164 Shouk (Yaseen and Shouk 2011) also reported that due to the lower ash, fat and protein content of 165 corn starch compared to semolina flour, increasing the replacement of flour caused a significant 166 decrease in ash, fat and protein content of the pasta. Similarly with these results, Mossadegh et al. 167 (Mossadegh, Tavakoli et al. 2018) and Crizel et al. (Crizel, Rios et al. 2015) observed that the 168 addition of potato fibre and orange fibre to the pasta formulation didn't significantly change the 169 fat content of the samples, however reduced the protein content.

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171 **Table 2.** The chemical composition of semolina flour, PS, TS and DKF used in this research.

Composition	Semolina flour	PS	TS	DKF
Moisture (%)	11.83 ± 0.37 °	$12.65 \pm 0.19$ <sup>b</sup>	13.93 ± 0.22 ª	$6.92 \pm 0.16$ <sup>d</sup>
Fat (%)	$1.16\pm0.03$ °	$1.28\pm0.01$ b	$0.41\pm0.03$ d	$3.14\pm0.05$ a
Protein (%)	$13.62 \pm 0.09$ <sup>a</sup>	$0.14\pm0.02$ $^{\rm c}$	$0.17\pm0.04$ $^{\rm c}$	$2.23\pm0.05$ $^{\rm b}$
Carbohydrate (%)	$57.80 \pm 0.41$ <sup>d</sup>	$85.52\pm0.47$ $^{\rm a}$	$83.44 \pm 0.50$ <sup>b</sup>	$79.94 \pm 0.38$ °
Total ash (%)	$0.74\pm0.03$ <sup>b</sup>	$0.12\pm0.02$ d	$0.23 \pm 0.02$ <sup>c</sup>	$1.02\pm0.05$ $^{\rm a}$
Fiber (%)	$3.46 \pm 0.27$ b	$0.61 \pm 0.08$ <sup>c</sup>	$0.36 \pm 0.11$ <sup>d</sup>	$73.94 \pm 0.46$ <sup>a</sup>

Values represent mean  $(n=3) \pm SD$ . Different letters in each row represent significant difference at 5% level of probability among samples. DKF: date kernel fiber, PS: potato starch, TS: tapioca starch.

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Table 3. Chemical composition of pasta treatments.

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Treatments	Moisture (%)	Fat (%)	Total ash (%)	Protein (%)
T0 (control)	$10.31 \pm 0.37$ <sup>a</sup>	$1.10\pm0.05$ <sup>a</sup>	$0.72\pm0.01$ a	$11.13 \pm 0.04$ <sup>a</sup>
T1	$10.83\pm0.24$ $^{\rm a}$	$1.13\pm0.03$ <sup>a</sup>	$0.64 \pm 0.01$ <sup>b</sup>	$0.95 \pm 0.07$ <sup>b</sup>
T2	$10.82 \pm 0.30$ <sup>a</sup>	$1.12\pm0.03$ <sup>a</sup>	$0.66 \pm 0.02$ <sup>b</sup>	$1.01 \pm 0.02$ <sup>b</sup>
T3	$10.81\pm0.27$ $^{\rm a}$	$1.10\pm0.04$ $^{\rm a}$	$0.62\pm0.02$ <sup>b</sup>	$0.97\pm0.05$ <sup>b</sup>
T4	$10.74 \pm 0.19$ <sup>a</sup>	$1.14\pm0.01$ $^{\rm a}$	$0.64\pm0.03$ <sup>b</sup>	$1.00\pm0.04$ <sup>b</sup>
T5	$10.62 \pm 0.35$ <sup>a</sup>	$1.13\pm0.03$ <sup>a</sup>	$0.64\pm0.02$ <sup>b</sup>	$0.95 \pm 0.06$ <sup>b</sup>
T6	$10.63 \pm 0.14$ <sup>a</sup>	$1.13 \pm 0.02$ <sup>a</sup>	$0.61\pm0.04$ <sup>b</sup>	$0.94\pm0.07$ <sup>b</sup>
Τ7	$10.60 \pm 0.21$ a	$1.14\pm0.02$ $^{\rm a}$	$0.63 \pm 0.02$ <sup>b</sup>	$1.04\pm0.08$ <sup>b</sup>
T8	$10.68\pm0.28$ $^{\rm a}$	$1.12\pm0.04$ $^{\rm a}$	$0.64\pm0.01$ <sup>b</sup>	$0.99 \pm 0.05$ <sup>b</sup>

179 Values represent mean  $(n=3) \pm$  SD. Different letters in each column represent significant difference at 5% level of 180 probability among samples.

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## 182 **3.2. Phenylalanine content of pasta**

183 PKU is a genetic disease caused by the deficiency of the phenylalanine hydroxylase enzyme, 184 which converts phenylalanine to tyrosine. This disease causes severe neurological complications 185 including irreversible mental disability (MacDonald, Van Wegberg et al. 2020). Therefore, the 186 level of protein and phenylalanine in the food of these patients is very vital and important. The 187 results of the examination the phenylalanine content of pasta (Figure 2) showed that the pasta 188 based on semolina flour (control) had the highest level of phenylalanine (530.58 mg/100 g dry 189 matter), and the complete replacement of wheat flour in the pasta formulation with different levels 190 of PS and TS caused a significant decrease in the amount of phenylalanine in the produced low-191 protein pastas (p < 0.05). However, there was no significant difference between the phenylalanine 192 content of different pasta treatments containing starches, and the phenylalanine content of these 193 treatments were in the range of 24.49-26.60 mg/100 g dry matter. The lower phenylalanine content 194 of the pasta compared to the control sample is due to the much lower protein content of PS and TS 195 compared to semolina flour. In this regard, Özboy (2002) stated that the starches contain very low 196 amounts of protein and phenylalanine and are therefore suitable for developing bakery products 197 for PKU patients. Yaseen et al. (Yaseen, Abd-El-Hafeez et al. 2011) reported that with the increase 198 of corn starch level in Egyptian bread formulation, the phenylalanine content of the samples 199 significantly decreased. Aghadadashzadeh Eslahi & Shams (Aghadadashzadeh Eslahi and Shams 200 2019) also showed in agreement with the results of this research that with the increase in the 201 replacement level of modified corn and tapioca starch in toast breads, their phenylalanine content 202 decreased significantly.

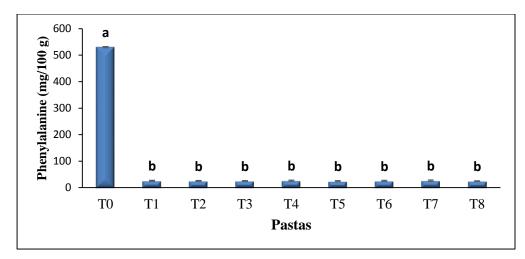




Figure 2. Phenylalanine content (mg/100 g) of pastas. Bars represent mean  $(n=3) \pm SD$ . Different letters on the bars indicate significant difference at 5% level of probability among samples.

#### 207 **3.3. Cooking loss of pastas**

208 The cooking process is a key process in determining the desired quality of pasta. During the 209 cooking process of pasta and as a result of the application of heat and water absorption, various 210 reactions occur, including starch gelatinization, protein swelling, and increase in volume and 211 weight of pasta. Cooking loss indicates the exit of solid materials during the pasta cooking process 212 and is a main indicator that is widely used to show the overall performance of the cooking, and 213 indicates the resistance to decomposition during pasta cooking (Larrosa, Lorenzo et al. 2016). 214 During cooking, less solids are extracted from the pasta, the higher the quality of the cooked pasta 215 (Del Nobile, Baiano et al. 2005). The cooking loss values of the pastas were measured and the 216 results are given in **Table 4**. The pasta based on semolina flour (control) had the lowest amount of 217 cooking loss, and the replacement of flour with the combination of PS and TS caused a significant 218 increase in the cooking loss percentage of the low-protein and pastas (p < 0.05). However, there 219 was no statistically significant difference between the cooking loss values of the pastas containing 220 starches, and the cooking loss values of these treatments were in the range of 8.10-8.59%. The 221 research has shown that flours with low protein and gluten-free have higher water absorption and 222 more cooking loss due to the absence of gluten network, because gluten reduces the absorption of 223 water by starch granules and prevents their leakage by preserving various compounds such as 224 gelatinized starches, carbohydrates and proteins (Yahyavi, Kamali Rousta et al. 2020). Hosseini 225 and Ardestani (Hosseini and Ardestani 2015) found protein reduction to be related to increase 226 cooking loss of pasta with protein removal. Overall, in gluten-free pasta, starch polymers are

227 effectively trapped in the matrix due to the absence of gluten, and the cooking loss of these 228 products is higher compared to conventional pasta (Marti and Pagani 2013, Cui, Zhao et al. 2020). 229 The maximum acceptable limit for pasta cooking loss is 10%, and cooking loss values less than 230 10% indicate good pasta quality (Bouasla, Wójtowicz et al. 2017). Since the cooking loss values 231 of all pastas produced in this research were less than 10%, so these treatments had good quality. 232 Hydrocolloids such as xanthan gum help create a stronger network in gluten-free products, which 233 traps starch granules and reduces cooking loss (Milde, Chigal et al. 2020). In the present study, 234 due to the use of xanthan gum in the pasta formulations as a texture improver, part of the increased 235 cooking loss due to the replacement of semolina flour with starches was compensated. Yaseen and 236 Shouk (Yaseen and Shouk 2011) reported that the cooking loss of control pasta was 10.6%, and 237 with the increase in the replacement of wheat flour with corn starch in the pastas, the cooking loss 238 increased and at the level of 70% replacement, the cooking loss reached 22.0%. Tao et al. (Tao, 239 Wang et al. 2020) also found that with the increase in the level of potato starch from 5% to 30% 240 in the noodle formulation, the cooking loss of the noodles increased.

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#### 242 **3.4. Cooking time of pastas**

243 During the cooking process of pasta in water, water absorption and starch granules gelatinization 244 occurs. As a result of applying heat, the gluten is denatured at the same time as starch is gelatinized. 245 However, gluten still retains a part of its water absorption ability (Palavecino, Bustos et al. 2017). 246 The best and most appropriate cooking time is when a large number of starch granules have 247 become gelatinized. The presence of gluten increases the cooking time of pastas by affecting the 248 temperature of starch gelatinization (Hosseini and Ardestani 2015). Cooking time is generally a 249 test that indicates the time required to cook pastas in boiling water and ready to eat (Milde, Chigal 250 et al. 2020). The results of examining the cooking time of pasta (Table 4) showed that pasta based 251 on semolina flour (control) had the longest cooking time (10.21 min), and the replacement of flour 252 with the combination of PS and TS led to a significant reduction in the cooking time of low-protein 253 pastas (p<0.05). By increasing the percentage of PS in the formulation, a decrease in the cooking 254 time of the pasta was observed (p < 0.05). The lowest cooking time was for the treatment containing 255 75% PS (7.93 min), but there was no significant difference between this treatment and the pasta 256 containing 65% PS+10% TS (8.04 min). The most effective factor in the effect of starches on the 257 pasta cooking time is the pasting temperature, and starches with a higher pasting temperature have

258 a higher resistance to the swelling and breaking of starch granules and increase the cooking time 259 (Sonia et al., 2019). The research has shown that the pasting temperature of PS (67.9 °C) is lower 260 than that of TS (71.6 °C) (Park, Kim et al. 2009, Sharma, Oberoi et al. 2009). Therefore, with the 261 increase of PS in pasta formulation, a greater decrease in the cooking time of the produced pastas 262 was observed. Adding hydrocolloids can increase the cooking time of pasta because hydrocolloids 263 limit the moisture content of the product and delay the swelling of starch granules (Kaur, Singh et 264 al. 2002). Sonia et al. (Sonia, Julianti et al. 2019) observed that adding xanthan gum increased the 265 cooking time of gluten-free pasta. Chillo et al. (Chillo, Laverse et al. 2007) reported that the 266 addition of pre-gelatinized corn starch and CMC to spaghetti reduced the cooking time by 3.5 267 times in samples based on amaranth flour compared to samples based on semolina flour. In this 268 research, the reduction of cooking time was attributed to the absence of gluten in pastas without 269 wheat flour. In fact, it seems that the absence of the gluten network facilitates the diffusion of 270 water through the product matrix and reduces the time required for water to reach the center of the 271 product during the cooking process.

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**Table 4.** Cooking loss and cooking time of pasta treatments.

Treatments	Cooking loss (%)	Cooking time (min)
T0 (control)	$6.78\pm0.40$ <sup>b</sup>	$10.21 \pm 0.14$ <sup>a</sup>
T1	$8.59 \pm 0.29$ <sup>a</sup>	$7.93 \pm 0.09 \ {\rm f}$
T2	$8.47\pm0.38$ a	$8.04\pm0.13~^{ef}$
T3	$8.41\pm0.26$ a	$8.25\pm0.17$ de
T4	$8.23 \pm 0.41$ a	$8.29 \pm 0.10^{\ d}$
T5	$8.10 \pm 0.24$ <sup>a</sup>	$8.35 \pm 0.14$ <sup>cd</sup>
T6	$8.19\pm0.31$ a	$8.40 \pm 0.16$ <sup>cd</sup>
T7	$8.29\pm0.46$ $^{\rm a}$	$8.56\pm0.10~^{bc}$
T8	$8.24\pm0.33$ a	$8.70\pm0.07~^{\rm b}$

Values represent mean (n=3) ± SD. Different letters in each column represent significant difference at 5% level of probability among samples.

### 3.5. Color of pasta

277 The color of pasta is one of the important parameters in product quality evaluation. Color is a 278 quality factor that affects consumer acceptance and product selection in the sale market (Xiong, 279 Chen et al. 2022). The color of raw pastas was determined using Hunterlab device and the results 280 are presented in Table 5. The control pasta had the lowest L\* (68.38) and the highest a\* (3.56) and 281 b\* (25.82), the color of low-protein and pasta containing PS and TS was brighter than the control 282 sample and less red and yellow. However, there was no statistically significant difference between 283 the color indexes of pastas containing starches, and the L\*, a\* and b\* values of these pasta samples 284 were in the range of 73.71-74.49, 0.76-0.86, and 20.30-20.79, respectively. The white color of PS 285 and TS is the reason for the increase in brightness of the color of gluten-free pastas compared to 286 the control. DKF also has a relatively white color. The researchers suggested that the ideal values 287 for the brightness index of the pasta color are greater than 60, and values less than 50 indicate the 288 overall darkness of the pasta (Luo, Wang et al. 2020). In agreement with the results of the present 289 study, Yaseen and Shouk (2011) also reported the lightening of the color of pasta with different 290 levels of corn starch compared to the control, and in their research, the intensity of redness and 291 yellowness of pasta containing corn starch was less than the control. The effect of xanthan gum in 292 brightening and yellowing the color of gluten-free pasta was also observed in the research of Milde, 293 Chigal et al. (2020).

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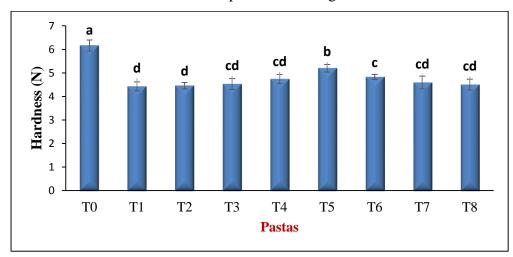
<b>Table 5.</b> Color indexes of raw pasta treatments.
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Treatments	L*	a*	b*
T0 (control)	$68.38 \pm 0.57$ <sup>b</sup>	$3.56 \pm 0.13$ a	$25.82 \pm 0.19$ <sup>a</sup>
T1	$74.49 \pm 0.47$ <sup>a</sup>	$0.78\pm0.04$ <sup>b</sup>	$20.46 \pm 0.30$ <sup>b</sup>
T2	$74.17 \pm 0.55$ <sup>a</sup>	$0.81\pm0.11$ b	$20.56 \pm 0.24$ <sup>b</sup>
T3	$74.22 \pm 0.51$ <sup>a</sup>	$0.84\pm0.08$ <sup>b</sup>	$20.41 \pm 0.27$ <sup>b</sup>
T4	$73.96 \pm 0.62$ <sup>a</sup>	$0.83\pm0.05$ <sup>b</sup>	$20.79 \pm 0.32$ <sup>b</sup>
T5	$73.71\pm0.38$ $^{\rm a}$	$0.86\pm0.09$ <sup>b</sup>	$20.75 \pm 0.17$ <sup>b</sup>
T6	$74.04 \pm 0.49$ <sup>a</sup>	$0.80\pm0.07$ <sup>b</sup>	$20.47 \pm 0.23$ <sup>b</sup>
Τ7	$73.85 \pm 0.56$ <sup>a</sup>	$0.76\pm0.13$ $^{\rm b}$	$20.59 \pm 0.29$ <sup>b</sup>
T8	$73.96 \pm 0.44$ <sup>a</sup>	$0.79\pm0.05$ <sup>b</sup>	$20.30 \pm 0.34$ <sup>b</sup>

Values represent mean (n=3) ± SD. Different letters in each column represent significant difference at 5% level of probability among samples.

#### **3.5. Hardness of pasta**

299 Pasta's texture qualities are a crucial factor in determining whether or not customers will 300 ultimately accept the product, and it is mainly considered as the most important qualitative aspect 301 of cooked pasta. In terms of customers' perspectives, high water absorption capacity, low cooking 302 loss and favorable texture (high hardness and low viscosity) can be considered as high cooking 303 quality (Luo, Wang et al. 2020, Zhang, Yu et al. 2023). The hardness of the pastas were examined 304 using a texture analyzer, and the results are shown in Figure 3. The control sample had the highest 305 hardness (6.16 N), and the hardness of low-protein and cooked pastas were significantly lower 306 than the control (p<0.05). Among the different low-protein pastas, the treatment containing 35% 307 PS+40% TS had the highest hardness. Overall, the hardness values of the pastas containing 308 starches were in the range of 4.43-5.20 N. The gluten present in wheat flour creates a strong 309 network in the dough and therefore the texture of the product containing gluten is desirable. So, 310 by removing gluten from the pasta formulation, the texture of the final product becomes more 311 fragile and its strength and hardness decrease (Yahyavi, Kamali Rousta et al. 2020). Hydrocolloids 312 are one of the most important additives used to improve the quality of gluten-free products, and 313 due to their gelling properties, they have a positive effect on the texture of gluten-free pasta and 314 improve the hardness of the texture (Chauhan, Saxena et al. 2017). In the present study, xanthan gum was used to compensate for some of the textural changes due to the removal of semolina flour 315 316 and gluten. Jung and Yoon (Jung and Yoon 2017) also reported that the hardness of pastas 317 containing different levels of buckwheat, acorn and mung bean starches was lower than the control 318 pasta. Milde et al. (Milde, Chigal et al. 2020) also showed the improvement of the hardness and 319 strength of gluten-free pastas based on corn flour and cassava starch due to the addition of xanthan 320 gum. These researchers stated that the hydrophilic compounds of hydrocolloids react with proteins 321 through ionic charges and improve the structure of pasta. Yaseen and Shouk (Yaseen and Shouk 322 2011, Wang, Guo et al. 2022) found that incorporating high levels of corn starch into a low-protein 323 pasta formulation resulted in a brittle batter prone to breakage.



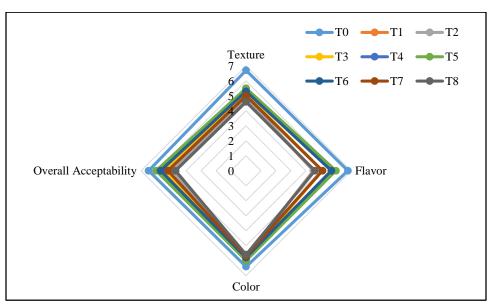
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Figure 3. Hardness values (N) of pastas. Bars represent mean  $(n=3) \pm SD$ . Different letters on the bars indicate significant difference at 5% level of probability among samples.

### **3.6.** Sensory evaluation of pastas

The sensory characteristics of cooked pastas, including texture, flavor, color and overall acceptability, were evaluated using a 7-points Hedonic scales, and the results are shown in Figure 4. The control sample got the highest sensory scores, because this sample is the common and commercial sample available in the market and its sensory characteristics are liked by consumers. Substitution of semolina flour in the formulation of gluten-free pastas with the combination of PS and TS caused a decrease in texture, flavor, color and overall acceptability scores of the produced pasta compared to the control (p<0.05). In terms of texture, flavor and overall acceptability, except 336 for the treatments containing 75% PS, 75% TS and the combination of 65% PS+10% TS, which 337 received an average score, the other treatments had scored in the good to very good range. Despite 338 the decrease in the color score of the pastas compared to the control, according to the results, the 339 light color created in the pastas due to the replacement of wheat flour with the combination of PS 340 and TS was desirable and acceptable to consumers. Among the different pastas, the pasta 341 containing 35% PS+40% TS got the best overall acceptance score (6.00), and it was the best 342 treatment in terms of sensory. Yaseen and Shouk (Yaseen and Shouk 2011) observed that by 343 increasing the level of corn starch in the low protein pasta formulation, the sensory scores of the 344 samples decreased, however, the substitution of corn starch up to 60% was sensory acceptable. Tao et al. (Tao, Wang et al. 2020) investigated the effect of different levels of PS on the sensory 345 346 properties of noodles and reported that noodles containing PS had high and favorable sensory 347 scores.



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Figure 4. Sensory characteristics of cooked pasta.

### **4. CONCLUSIONS**

The results of this research showed that replacing semolina flour with the combination of PS and TS significantly reduced the ash, protein and phenylalanine content of the pastas. The cooking properties of pastas was also affected by the substitution of wheat flour with the combination of starches, so that the cooking loss of low-protein pastas were higher than the control sample, but they have less cooking time. Increasing the level of PS in the formulation also caused a further decrease in cooking time. The color of low-protein and pastas was lighter than the control, but their redness and yellowness was less. The replacement of wheat flour with the starches also made the texture of the pastas softer compared to control. The results of this study indicated that by using the combination of PS and TS instead of semolina flour, as well as xanthan gum and DKF, pasta with low phenylalanine can be produced for patients with PKU, and the produced pastas are also suitable and usable for celiac patients because they don't contain gluten. The best treatment in this research was the pasta containing 35% PS+40% TS, which had the best texture and the highest sensory scores.

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# تولید پاستای عملگرا با فنیل آلانین کم بر پایه نشاسته سیب زمینی و تاپیوکا

- 483
- عليرضا رضايي، مانيا صالحي فر، و وجيه فدايي نوغاني 484
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#### چکیدہ

فنیل کتونوری (PKU) یک اختلال ژنتیکی است که نیاز به رژیم غذایی کم پروتئین و فنیل آلانین دارد. این مطالعه با هدف 486 487 بررسی امکان سنجی تولید یاستای عملگرا کم پروتئین با استفاده از نشاسته سیب زمینی و تاییوکا انجام شد. در فرمول یاستا، آرد سمولینا با مخلوطی از نشاسته سیب زمینی و تاییوکا جایگزین شد. فیبر هسته خرما و صمغ زانتان به ترتیب به 488 عنوان ترکیبات پری بیوتیک و تقویت کننده بافت ترکیب شدند. خواص فیزیکوشیمیایی (رطوبت، چربی، خاکستر کل، 489 490 پروتئین، فنیل آلانین، از دست دادن پخت، زمان پخت، شاخص های رنگ و سختی) و ویژگی های حسی (بافت، طعم، رنگ و مقبولیت کلی) با نمونه کنترل (بر اساس آرد سمولینا) مقایسه شدند. نتایج نشان داد که تغییر معنی داری در رطوبت و 491 محتوای چربی پس از جایگزینی مشاهده نشد، اما کاهش معنی داری در محتوای خاکستر و پروتئین مشاهده شد (p). 0/05-492 493 در نتیجه، سطح فنیل آلانین از 530.58 میلی گرم در 100 گرم در نمونه شاهد به 24.49-26.60 میلی گرم در 100 گرم در ياستا عملگرا كاهش يافت. جايگزيني آرد با نشاسته باعث افزايش تلفات يخت، كاهش زمان يخت و كاهش سختي ياستا 494 495 نسبت به شاهد شد (p). 0/05-پاستا حاصله \*L بالاتر و مقادیر \*a و \*b کمتری نسبت به شاهد نشان داد. ارزیابی حسی 496 نشان داد که پاستا حاوی 35 درصد نشاسته سیب زمینی و 40 درصد نشاسته تاپیوکا بالاترین امتیاز را به دست آورد که 497 نشان دهنده مقبولیت آن است. به طور کلی، این مطالعه نشان می دهد که ترکیب نشاسته سیب زمینی و تاپیوکا، همراه با فيبر هسته خرما و صمغ زانتان، توليد پاستا عملگرا و كم پروتئين مناسب براى بيماران PKU را امكان پذير مى كند. 498

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