

Production of low-fat frankfurter sausage using a new fat replacer: reconstituted agar hydrogel

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Abstract

There is a growing trend towards healthy meat products containing lower fat content. The use of fat replacers can solve the quality problems of low-fat products. This study aimed to produce low-fat frankfurter sausage using a new fat replacer. Thus, in the first stage, reconstituted agar hydrogel (RAH) was produced by adding water to agar cryogel after homogenization. In the second stage, RAH was used as a fat substitute in frankfurter sausage at 0, 25, 50, 75, and 100%, and the characteristics of the resulting low-fat sausage, including chemical composition, water holding capacity, cooking loss, texture characteristics, porosity, color, percentage of fat after frying, oil absorption, pH over time and sensory evaluation were examined. Results showed that RAH could form two kinds of gel by temperature changes: a low-set gel at 55°C and a high-set gel at 90°C. Substitution of oil in the sausages caused an increase in moisture content, porosity, oil absorption, and cooking loss. On the other hand, this replacement reduced fat content, cutting force, water holding capacity, fat percentage after frying, and texture properties such as hardness, cohesiveness, springiness, gumminess, and chewiness. The results of pH measurements over time showed that sulfated antimicrobial groups in the agar structure delayed the spoilage of sausages containing RAH compared to the control sample. Sensory evaluation showed that RAH-containing sausages were not significantly different from the control sample in terms of color, juiciness, and texture. However, flavor and overall acceptance increased significantly under the influence of this substitution ($P < 0.05$). So, consumers selected a sample with 25% replacement as the optimal sample. As a result, RAH can be successfully used as a fat replacer in low-fat products with desirable quality characteristics.

Keywords: Fat substitute, Cryogel, Reconstituted hydrogel, Low-fat sausage.

Introduction

Frankfurters are emulsion-type sausages that are widely consumed. In restructured meat products, fat plays an important role in stabilizing meat emulsions, reducing cooking loss, improving water holding capacity, and providing organoleptic quality (Rather et al. 2015). It also provides essential nutrients such as fat-soluble vitamins and essential fatty acids and plays an important role in the flavor and texture of meat products. On the other hand, a high intake of saturated fats increases the risk of obesity and cancer. It is also closely linked to high blood cholesterol and coronary heart disease.(Rather et al. 2015; Safa et al. 2015; Shin et al. 2022; Souza et al. 2019). With an awareness of their nutrition and health, consumers today prefer to eat healthier meat products with fewer calories. From a nutritional point of view, it is desirable to reduce the high amount of fat in meat products. But, the elimination or reduction of fats in these products reduces the sensory quality of the final product and mainly affects its texture (Chen et al. 2021; Y. Kumar 2021; Pietrasik and Soladoye 2021). In this regard, manufacturers have introduced several changes in an effort to compensate for the negative effects of reducing fat levels in reconstructed meat products. These modifications include the use of fat substitutes, which can be used to produce low-fat meat products with the same characteristics of high-fat meat products in terms of appearance and taste. FDA-approved fat replacers include carbohydrate-based, protein-based, and fat-based fat replacers (Y. Kumar 2021).

Hydrogels are three-dimensional networks of homopolymers or copolymers whose hydrophilic structure enables them to hold large amounts of water in their three-dimensional network. Softness, elasticity, swelling, non-dissolution in water, absorbent nature, flexibility (due to the presence of a lot of water in the structure), and the ability to store water are important characteristics of hydrogels. Natural hydrocolloids such as agar, alginate, gellan, guar, tragacanth, and pectin are natural polymers used to prepare hydrogels (Klein and Poverenov 2020; Nayak and Das 2018; Ahmed 2015). Agar, among these, is widely used in the food industry. Agar forms a gel with a high hydration capacity and can lose a lot of water without changing the gel structure. It can maintain its gel at high temperatures. It is considered the strongest gel-forming substance. Agar has sufficient resistance to hydrolysis to such an extent that in meat preservatives that require

sterilization at a temperature higher than 121 C, treatment can be done successfully without hydrolysis of agar (Phillips and Williams 2009).

So far, carbohydrate-based fat replacers have been widely used in meat products. For example, Rather et al.,(2015) used xanthan as a fat replacer in goshtaba. It was observed that oxidative stability and product quality of goshtaba increased with this hydrocolloid due to the antioxidant activity of xanthan. Also, hardness and taste intensity decreased with increasing xanthan concentration. Rather et al., (2016) investigated the effect of guar gum as a fat replacer on some quality parameters of the mutton product. The results of this experiment were reduced lipid oxidation, increased water holding capacity, reduced hardness and flavor intensity, and increased chewiness due to reduced fat levels. Abbasi et al., (2019) used tragacanth gum as a fat replacer in sausages. Delays in the formation of reactive oxidation materials (thiobarbituric acid), low carbonyl content at the end of storage, and thus increased oxidative stability and improved texture quality were some of the results of this replacement. Öztürk-Kerimoğlu (2021) used a gel system comprising pea protein and agar (PAG) as fat substitutes in meat emulsions. Fat replacement reduced total fat content while increased moisture, protein, and dietary fiber content. Emulsion stability and cook yield increased with increasing replacement rate. The lightness and yellowness of the modified samples were higher than the control samples, but their redness was lower than the control samples. Complete fat replacement with PAG gave a softer but more adhesive texture. In general, this replacement had favorable effects on the functional properties of meat emulsions. Jayarathna et al., (2022) investigated the effect of replacing vegetable oil with garlic inulin on the quality characteristics of chicken sausages. Sausages containing inulin had a higher score of taste and overall acceptance. The ash content, moisture, and protein content of sausages increased with increasing inulin levels. In contrast, the fat content decreased from 13.67% to 4.47% in products containing 3% inulin. Also, products containing 2% inulin had lower brightness than the control sample. As a result, garlic inulin can be successfully used as a fat substitute in sausages without changing the quality parameters of the meat.

Since the consumption of low-fat meat products has received a lot of attention from consumers, this study considers using a new form of agar hydrocolloid as a fat replacer. For this purpose, the production of reconstituted agar hydrogel from its cryogel, which is not previously used in any other studies, and replacing sausage fat with produced RAH will be evaluated.

Materials and methods

Production of reconstituted agar hydrogel

Agar solution (1%. w/v) was prepared by adding agar powder to distilled water at 45 °C gradually. Then, the solution was stirred for one hour at 95 °C until the agar was completely hydrated. The agar solution was immediately aerated with a homogenizer (T-25, IKA, Germany) at 11000 rpm for 15 min and then was cooled to 45 °C to form the agar gel (Ellis et al. 2017). After the gel formation, it was stored at -18 °C for 24 h and then dried by freeze-drying method to form agar cryogel. Water was added to agar cryogel (90%, w/w), and the mixture was homogenized at 11000 rpm for 3 min to obtain a solid fat-like texture called reconstituted agar hydrogel (RAH). The RAH was kept in a refrigerator (4°C) after production until sausage production (less than 24H).

Characterization of reconstituted agar hydrogel

The microstructure of agar cryogel was investigated by obtaining electron images from the surface and cross-section of agar cryogel using a field emission-scanning electron microscope (FE-SEM) (Quanta FEG-450, FEI, USA). Agar cryogel was broken into liquid nitrogen to prepare a cross-section of the sample. Then, the surface and cross-section of the agar cryogel were covered with gold and imaged at a voltage of 20 kV (Fayaz et al. 2017; Plazzotta et al. 2019).

To determine water holding capacity (WHC), Eppendorf weight without (a) and with RAH (b) was recorded. Then, the Eppendorf was centrifuged at 10000 rpm for 15 min. Afterward, the Eppendorf was turned upside down on filter paper for 3 min to remove separated water from RAH, and the Eppendorf was weighed again (c). WHC of the RAH was calculated using the following equations (Fayaz et al. 2017)

$$\text{Water migration (\%)} = (b-a) - (c-a) / (b-a) \quad (1)$$

$$\text{Water holding capacity (\%)} = 100 - \text{water migration} \quad (2)$$

To determine WHC at different temperatures, RAH samples were stored at 30, 60, and 90 °C for 45 min before WHC determination.

The back extrusion method was applied to determine the hardness of the RAH tissue. A texture analyzer device (SDM1, Santam, Iran) equipped with a 100 kg cell at a speed of 100 mm/min was used. The maximum force required to penetrate the RAH tissue at a depth of 6 mm was determined (Giacintucci et al. 2018). The texture hardness of solid fat was also measured.

RAH color was evaluated using a HunterLab colorimeter (ZE6000, Nippon Denshoko, Japan) based on the L*, a*, and b* color system. The color of solid fat was also determined.

Preparation of low-fat sausages using reconstituted agar hydrogel

Chicken meat (25% chicken breast and 75% chicken thigh) and other ingredients were obtained from a local market. According to a commercial sample formulation, the required ingredients were mixed in a food processor (FP3010, Brown, Germany) according to Table 1 to produce control frankfurter sausage. First, chicken meat, grated bell pepper and garlic were mixed for 3 min. Then, salt, phosphate, and crushed ice were mixed for 1 min. Sodium nitrite dissolved in cold water and crushed ice were added to the cutter. Soy protein isolate, powdered milk, and crushed ice were then mixed for 1 min. Finally, gluten, oil, wheat flour, starch, crushed ice, spices, and ascorbic acid were added in order. Then, the mixture was filled in polyamide coating with a diameter of 5 cm and a thickness of 0.1 mm and placed in boiling water for one hour until the center temperature of the sausage reached 72-75 °C. After cooking, the samples were cooled to room temperature (25°C) and kept in the refrigerator (4°C) for seven days. Low-fat sausages were produced by replacing the oil in the sausage formulation with RAH at 25, 50, 75, and 100% replacement.

Characterization of low- fat sausages

Chemical composition

The chemical composition of control and low-fat sausages, including moisture, fat, protein, and ash, were measured one day after production by freeze-drying, Soxhlet, Kjeldahl, and dry ashing methods, respectively, according to AOAC standard (2005) (Horwitz and Latimer 2005).

Water holding capacity (WHC)

Four pieces of filter paper with dimensions of $2.5 \times 2.5 \text{ cm}^2$ were folded and placed in an Eppendorf tube and weighed (w_1). The sausage sample (w_3) was placed in it and centrifuged at 14000 rpm for 10 min. After the sample was separated from the Eppendorf, the filter paper and Eppendorf tube were weighed again (w_2). WHC was calculated according to the following equation (Santana et al. 2015) :

$$\text{WHC (\%)} = 100 - ((w_2 - w_1) * 100 / w_3) \quad (3)$$

Cooking loss

Circular pieces of sausage with a thickness of 1 cm were cut and weighed (w_1). Then, they were deep-fat fried for 30 s. The surface oil of the sausages was removed using filter paper, and the sausage samples were weighed again after cooling (w_2). Cooking loss was calculated as follows (Lee and Chin 2016):

$$\text{Cooking loss (\%)} = (w_1 - w_2) / w_1 \quad (4)$$

Texture analysis

Sausage samples were cut with a diameter of 1.37 cm and a height of 1.6 cm with a v-shaped blade to perform the cutting test. Then, they were tested with a Warner-Bratzler blade at a constant speed of 100 mm/min using a texture analysis device (SDM1, Centam, Iran) (Abbasi et al. 2019).

Texture profile analysis (TPA) of different sausages was also evaluated using a texture analysis device (Instron, America) at 25 °C. First, the sausages were cut into cylinders with diameters and heights of 1.37 and 1.6 cm, respectively. Then, a stainless-steel cylindrical probe with a diameter of 3 cm was used for TPA testing. Compression of 50%, a number of cycles of 2 pcs, load cell of 100 N, speed of 50 mm/min, and trigger force of 0.07 N were applied to perform this test. The parameters of hardness, cohesiveness, springiness, gumminess, and chewiness were extracted from the obtained profiles (Fernández-López et al. 2006; Kouzounis et al. 2017; Surasani et al. 2022).

Oil absorption

Sausage samples were fried according to the previous section method. The fat content of raw (F_1) and fried sausage samples (F_2) was measured by the AOAC method (2005) using the following equation:

$$\text{Oil absorption (\%)} = (F_2 - F_1) / F_1 \quad (5)$$

pH over time

Sausage (2g) was mixed with distilled water (8 ml) and homogenized at 11000 rpm for one min. The pH of the samples was determined at intervals of 0.5, 3, 5, 9, 12, and 16 days after production using a manual pH meter (Acton, Malaysia) (Lee and Chin 2016).

Sensory evaluation

Control sausage and low-fat sausages with 25 and 50 % RAH as fat replacers were identified with three-digit codes and were simultaneously presented to 25 evaluators from different age

groups. The evaluators evaluated parameters of juiciness, flavor, texture, color, and overall evaluation based on seven-point hedonic scales, where the 7 number indicated a very good sample and the 1 number indicated a weak sample.(de Souza Paglarini et al. 2021; Gómez-Estaca et al. 2019; M. Kumar and Sharma 2004).

Results and discussion

Characterization of agar cryogel

Surface and cross-section images of agar cryogel are shown in Figure 1a and b, respectively, to evaluate its microstructure using scanning electron microscopy. Figure 1b well shows the porous network structure of agar cryogel. Hydrogen bonds solely cause the formation of the agar gel network. This gel network loses its water rapidly due to the sublimation during the drying process but retains its network structure, which is well seen in Figure 1b and was confirmed by the report of Patel *et al.*(2013) in the agar cryogel structure that shows the intensity of the hydrogen bonds present in the network. The gel structure of agar is intimately connected with the intensity of hydrogen bonds. The ability of agar to form a stable gel relies on the establishment of a network of hydrogen bonds between its molecular components. This is the same network that can trap water and cause the creation of hydrogel, confirming the high capacity of water storage.

Characterization of reconstituted agar hydrogel

The WHC of the reconstituted hydrogel prepared from agar cryogel was $99.39 \pm 0.11\%$. The high WHC of RAH is due to the agar structure, which can retain a high amount of water in its structure. The porous network structure of agar cryogel, shown in Figure 1, confirms the high-water holding capacity of RAH. The results of WHC of RAH samples at 30, 60, and 90 °C were 99.65 ± 0.17 , 92.1 ± 1.33 , and 99.38 ± 0.11 , respectively. The water holding capacity of the RAH decreased significantly with the temperature increasing to 60 °C. However, it increased significantly again as the temperature rose to 90 °C ($p < 0.05$). The agar gel network is created by only weak hydrogen bonds retaining water in its network. Therefore, the RAH network is weakened by temperature increasing up to 60 °C due to the weakening of hydrogen bonds. So, the WHC of the reconstituted hydrogel at 60 °C was significantly reduced. A very interesting and different phenomenon happened to the RAH compared to the agar gel observed during this test. The RAH turned into a gel at 90 °C, and the water was entrapped entirely into the structure of the reconstituted hydrogel.

Thus, the water holding capacity of RAH increased. It seems that the process of converting agar gel to cryogel has caused changes in the physical structure of the agar, providing hydrophobic interaction between RAH chains, which results in the formation of gel at high temperatures. Therefore, it can be said that RAH has a different property compared to agar powder, which is the ability of RAH to form two kinds of gel by temperature changes: 1) a low-set gel, which is thermoreversible, forms by cooling the RAH solution heated up to about 55 °C, and 2) a high-set gel, which is irreversible, forms by heating RAH solution up to 90 °C. This phenomenon is similar to the unique property of curdlan, which is not seen in other hydrocolloids. Curdlan can also form two types of gels: low-set gel (forms at about 55 °C) and high-set gel (forms at about 80 °C). Hydrophobic reactions between curdlan molecules at high temperatures cause a network structure to hold water inside (Phillips and Williams 2009).

Texture analysis of RAH was obtained by the back extrusion method at a depth of 6 mm. Results showed that the maximum force applied on the RAH was 4.00 ± 0.28 N. The hardness of the solid fat, measured by the same method, was 2.2 N. Comparison of the hardness of RAH with solid fat helps estimate the hardness of RAH. The similarity of these two numbers also indicates the possibility of using RAH as a substitution for solid fats in low-fat products.

The color parameters L^* , a^* , and b^* of the RAH were 54.83, -1.08, and 9.11, respectively. Results shows that the RAH sample has low lightness. Since homogenization is used to prepare RAH, the entry of a large volume of air into the reconstituted hydrogel network causes its color to darken. The negative value of a^* parameter in the RAH sample indicates a slight green tint of the RAH sample. The b^* parameter of RAH shows the yellowness of reconstituted hydrogel. The color of RAH was very similar to solid fat ($L^*=58.24$, $a^*=-2.42$, and $b^*=13.22$), which is an advantage of using RAH as a fat substitution in low-fat foods.

Characterization of low- fat sausages

Chemical composition and WHC

The chemical composition of prepared sausages containing different levels of RAH as fat replacers is shown in Table 2. The moisture content increased significantly with the replacement of sausage oil with RAH due to high water in the RAH structure. It is attributed to the ability of agar to retain much water in its structure during the preparation and curing process (Phillips and Williams 2009). Rather et al., (2015) reported that reconstituted meat products containing xanthan as a fat replacer

had more moisture content than the control sample due to reduced fat content and increased hydrocolloid content in the formulation. The fat content of sausage samples decreased with increasing the replacement ratio of oil with RAH. Rather et al., (2015) also reported that the fat content decreased in meat formulations containing xanthan as a fat replacer. In general, the use of hydrocolloids in different types of sausages effectively has reduced the fat content of products (Berizi et al. 2017; Glisic et al. 2019; Jayarathna et al. 2022; Kim et al. 2019; Souza et al. 2019). The fat content of samples with 100% replacing oil with RAH was not zero due to the fat content of chicken (2.9%) used in the formulation. The results obtained from the ash and protein contents showed that the sausages containing RAH and the control sample were not significantly different in terms of ash and protein contents ($p < 0.05$). Because agar does not contain protein. Also, agar ash content did not affect the ash content of the final sausage because only one percent of agar was used in the RAH making. Similarly, Salajegheh et al. (2018) showed that fat reduction did not significantly affect the level of ash and protein in sausage.

The results of Table 2 show no significant difference in WHC between the control sample and sausages containing 25 and 50% RAH, but the WHC of samples significantly reduced when the replacement increased to 75 and 100%. Agar is a hydrocolloid and has a high-water absorption and storage capacity. Therefore, it was expected that the WHC of the samples did not change in the low amount of replacing the fat with RAH (25 and 50%). However, the amount of water in the samples containing 75 and 100 % oil replacement with RAH was very high (Table 2). Therefore, the amount of protein available was insufficient to create a suitable emulsion holding a high volume of water. Zhu et al. (2023) used oleogel based on flaxseed gum/arabic gum as a fat substitute in sausage. They observed that the WHC of the control group was lower than the sausage samples containing 25 and 50% oleogel replacement. They stated that a higher WHC is beneficial for retaining water molecules in the gel structure and reducing the cooking loss of sausages.

Cooking loss

The results of Figure 2 show that the cooking loss of the samples increased with increasing the replacement ratio. WHC is one of the most important factors affecting the cooking loss of the sausages. The lower the WHC, the higher the cooking loss due to the higher water evaporation from the product during frying. On the other hand, reducing the amount of oil in sausages increases the moisture content as the emulsion cannot be fully formed, and as a result, the cooking loss

increases (Salajegheh et al. 2018). The water content of sausage samples increased by increasing the oil replacement with RAH (Table 2). So, the water of samples containing higher amounts of water evaporated more during frying, which increased the porosity of the fried samples and the cooking loss. Jiménez-Colmenero et al. (2010) added a combination of marine spaghetti gel/kongac to frankfurters to reduce fat and salt. It was observed that this combination increased cooking loss in gel/emulsion systems. These researchers attributed this effect to the early melting of the hydrocolloids and higher water release from the konjac gel during baking to 68°C. Dong Kim et al. (2019) also reported that adding konjac gel to low-fat sausage decreased the cooking loss of low-fat sausage but still had more cooking loss than normal-fat sausage.

Texture analysis

The results of TPA of sausages containing different amounts of RAH are shown in Table 2. The hardness of the samples decreased with increasing the percentage of RAH in the samples, which may be due to the increase of moisture content of sausages after replacing oil with RAH. The springiness and cohesiveness also decreased with increasing the replacement amount of RAH. The fat content is one of the many factors affecting the springiness and cohesiveness of the sausages. The lower the fat content of the sample, the lower these parameters. Therefore, the reduction of these texture parameters can be due to the decrease in fat content (Mendoza et al. 2001). The gumminess was another texture parameter that decreased with increasing the content of RAH in the sausages due to the reduction of hardness and cohesiveness. Chewiness reduction after replacing oil with RAH was due to the decrease in gumminess (Fernández-López et al. 2006).

The result of the cutting test is also shown in Table 2, which indicates that the higher the amount of RAH in the sausages, the lower the force required to cut the samples and, thus, the higher the cutting ability. The cohesiveness and hardness of the sausages decreased after replacing oil with RAH, which can reduce the force required to cut the samples. On the other hand, reducing the amount of fat causes the protein structure not to open completely during the preparation of sausages, which will create weaker gels and thus increase the cutting ability of the samples. Zhu et al.(2023) examined the textural characteristics of sausages formulated with the replacement of pork back fat by flaxseed gum/arabic gum oleogel. It was observed that the hardness of sausages formulated with oleogels was higher compared to the control group. But the hardness of emulsified sausages decreased when the replacement ratio of fat with oleogels increased from 25% to 100%. The cohesiveness and chewiness of sausages showed a similar trend to hardness. However, there

was no significant difference in springiness between the control group and the sausage emulsified with oleogel.

Oil absorption and fat content after frying

The oil absorption of sausages containing 0, 25, and 50 % RAH was 16.35 ± 1.12 , 45.87 ± 5.36 , and 46.48 ± 4.33 , respectively. Results show a significant increase in oil absorption of sausages after replacing oil with RAH. However, there is no significant difference between the oil absorption of samples containing 25 and 50% RAH. The results of WHC showed that replacing oil with RAH reduced the WHC of the samples, which could be a good reason for the oil absorption increase. As higher amounts of water evaporate during the frying in samples with lower WHC and cause higher porosity, higher amounts of oil will absorb in the sausages.

After frying, the fat content of sausages was 22.51 ± 0.5 , 21.88 ± 1.01 , and 16.41 ± 0.7 in sausages containing 0, 25, and 50% RAH, respectively. The results show that oil replacement with RAH significantly affected the fat content of the samples after frying ($p < 0.05$). The fat content of the samples after frying was significantly lower in the samples containing RAH as a fat replacer. In the control sample, which is high in fat, the fat content can be expected to increase after frying; however, the amount of fat after frying did not decrease significantly in the samples containing 25% RAH, which is due to the higher oil absorption of this sample compared to the control sample. The results of this test show that although the oil absorption of samples containing RAH increased; however, due to the less oil used in the formulation of low-fat sausages, the fat content of the final sample in low-fat sausages is less than the control sample, which shows the positive effect of using RAH on fat reduction in low-fat products. These results were consistent with the findings of Zhu et al. (2023), who used flaxseed gum-based oleogel as a fat substitute in emulsion sausages. They observed that increasing the concentration of flaxseed gum increased the rate of oil loss significantly due to the tighter network structure of the oleogels.

pH over time

The pH during storage of sausages prepared with different percentages of oil replacement with RAH is presented in Table 3. The pH of all three samples decreased over time. However, this decrease was not significant until the ninth day ($p < 0.05$). While it significantly decreased until the 16th day due to the microbial spoilage over time with lactic acid bacteria, which are the dominant microorganisms in meat products. The pH difference between the three samples was not

significant until the ninth day, which shows that replacing sausage oil with RAH did not cause the samples to become more perishable. Interestingly, from the ninth to the twelfth day, when the pH decreased in all three samples, the intensity of this decrease was higher in the control sample than in the samples containing RAH. Therefore, the results show that replacing sausage oil with RAH slowed down the spoilage of the samples. It seems that the presence of low amounts of sulfate in the agarose structure of agar acts as an antimicrobial agent and slows down the spoilage of sausages containing agar compared to the control sample (Liu et al. 2017).

Sensory evaluation

The sensory evaluation of sausages prepared with different amounts of RAH is presented in Table 4. The color, juiciness, and texture of sausages containing RAH were not significantly different from the control samples. However, there was a significant difference between the samples in terms of aroma and taste and overall acceptance ($p < 0.05$). Interestingly, the aroma and taste of the sample containing 25% RAH were significantly more desirable than those of the control sample. Fat-soluble flavoring agents seem well sensed in the control sample due to the high amounts of oil in the formulation. However, water-soluble flavoring agents are not released well, so many flavors do not feel good in the control. However, the total fat-soluble and water-soluble flavors are well released and sensed in the sample containing 25% RAH due to the desirable balance between hydrophobicity and hydrophilicity of the sample, so the flavor improves at this percentage of replacement. Finally, the overall evaluation shows that the samples containing 25% RAH and control samples are similar. The sample containing 50% RAH, still from the consumer's point of view, had no significant difference from the control sample in terms of overall acceptance. Kim et al. (2019) used konjac gel and three herbal powders (aloe vera, cactus pear, or wheat germ) as pork fat substitutes to prepare low-fat frankfurter sausages. It was observed that cactus pear konjac gel can be used as a frankfurter fat substitute with a positive effect on color, taste, smell, and overall acceptability.

Conclusions

Considering the increasing awareness and concern of consumers regarding the harm of high-fat and high-calorie meat products that cause the spread of some diseases, using a way to reduce the amount of fat helps to overcome these concerns. On the other hand, eliminating or reducing the

fats in the product reduces the quality of sensory properties in the final product and especially affects its texture. The use of fat substitutes can be a way to solve the quality problems of these products. Regenerated agar hydrogel (RAH) was used instead of oil to prepare frankfurter sausage to achieve this goal. Using RAH instead of oil positively affected the chemical compounds, including moisture and fat, texture, color, and sensory evaluation of sausage samples. Also, due to sulfated antimicrobial groups in the agar structure, the spoilage occurred later in the sausage samples containing RAH than in the control sample. Therefore, RAH can be used in meat products to reduce the amount of fat and, at the same time, maintain the quality characteristics of the product. Sensory evaluators gave better scores to the sample with 25% RAH. Finally, sausages with two percent replacement rates of 25 and 50 % can be introduced as the best low-fat sausages.

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Table 1. Formulation of different sausages prepared with RAH replacer.

Ingredient	Percentage of oil replacement with RAH				
	0	25	50	75	100
Chicken meat	45	45	45	45	45
Sunflower oil	18	13.5	9	4.5	0
RAH	0	4.5	9	13.5	18
Salt (NaCl)	1.7	1.7	1.7	1.7	1.7
Sodiumpolyphosphate	0.57	0.57	0.57	0.57	0.57
Sodium nitrite	0.01	0.01	0.01	0.01	0.01
Ascorbic acid	0.02	0.02	0.02	0.02	0.02
Soy protein isolate	0.95	0.95	0.95	0.95	0.95
Gluten	1.12	1.12	1.12	1.12	1.12
Wheat starch	1.86	1.86	1.86	1.86	1.86
Milk powder	1.07	1.07	1.07	1.07	1.07
Wheat flour	4.95	4.95	4.95	4.95	4.95
Garlic	1.15	1.15	1.15	1.15	1.15
Sugar	0.28	0.28	0.28	0.28	0.28
Ice	20	20	20	20	20
Bell pepper	2	2	2	2	2
Spices*	1.32	1.32	1.32	1.32	1.32

*Spices contained pepper, nutmeg, ginger, coriander seeds, thyme, chicken flavor, liquid smoke, and pepper flavor.

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Table 2. Chemical composition, WHC, and texture analysis of sausages containing different amounts of RAH.

Parameter	RAH amounts (%)				
	0	25	50	75	100
Moisture (%)	59.00± 1.47 ^e	62.50 ± 0.70 ^d	65.30 ± 0.23 ^c	66.70 ± 0.19 ^b	67.90 ± 0.14 ^a
Fat (%)	19.30 ± 0.35 ^a	15.00 ± 0.14 ^b	11.20 ± 0.14 ^c	6.40 ± 0.28 ^d	1.30 ± 0.14 ^e
Protein (%)	12.90 ± 0.33 ^a	12.20 ± 0.50 ^a	12.10 ± 0.87 ^a	12.40 ± 0.38 ^a	12.09 ± 0.57 ^a
Ash (%)	2.99 ± 0.15 ^a	3.19 ± 0.16 ^a	3.46 ± 0.48 ^a	3.38 ± 0.53 ^a	3.39 ± 0.09 ^a
WHC (%)	77.65 ± 2.16 ^a	76.31 ± 1.21 ^a	76.36 ± 0.81 ^a	70.83 ± 2.54 ^b	67.38 ± 1.75 ^c
Hardness (kg)	17.23 ± 1.46 ^a	11.86 ± 0.92 ^b	12.06 ± 0.24 ^b	9.42 ± 0.80 ^c	9.75 ± 0.32 ^c
Springiness (cm)	6.83 ± 0.14 ^a	6.51 ± 0.27 ^b	5.83 ± 0.30 ^c	5.60 ± 0.22 ^{cd}	5.34 ± 0.14 ^d
Cohesiveness	0.58 ± 0.07 ^a	0.55 ± 0.03 ^a	0.50 ± 0.04 ^b	0.41 ± 0.01 ^c	0.34 ± 0.01 ^d
Gumminess (kg)	10.13 ± 1.37 ^a	6.36 ± 0.43 ^b	6.15 ± 0.62 ^b	3.94 ± 0.31 ^c	3.38 ± 0.19 ^c
Chewiness	69.28 ± 9.47 ^a	43.17 ± 2.57 ^b	35.86 ± 3.28 ^c	22.12 ± 1.37 ^d	18.10 ± 1.19 ^d
Cutting test	2.83 ± 0.46 ^a	1.95 ± 0.36 ^b	1.80 ± 0.25 ^b	1.26 ± 0.18 ^c	1.21 ± 0.15 ^c

Lowercase letters in each row indicate a significant difference between the samples (p <0.05)

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Table 3. pH of sausages containing different amounts of RAH during storage.

Time (day)	RAH amounts (%)		
	0	25	50
0.5	6.12 ± 0.015 ^{a-c}	6.11 ± 0.004 ^{a-c}	6.10 ± 0.014 ^b
3	6.12 ± 0.003 ^{a-c}	6.12 ± 0.003 ^{a-c}	6.13 ± 0.004 ^a
5	6.12 ± 0.007 ^{a-c}	6.13 ± 0.006 ^{a-c}	6.14 ± 0.006 ^a
9	6.07 ± 0.023 ^c	6.08 ± 0.009 ^b	6.12 ± 0.009 ^{a-c}
12	5.22 ± 0.049 ^f	5.81 ± 0.072 ^d	6.17 ± 0.042 ^a
16	5.18 ± 0.03 ^f	5.64 ± 0.11 ^e	5.6 ± 0.022 ^e

Lowercase letters indicate a significant difference between samples (p <0.05).

Table 4. Sensory evaluation of sausages containing different amounts of RAH.

Test	RAH amounts (%)		
	0	25	50
Color	6.6 ± 0.57 ^a	6.36 ± 0.9 ^a	6.24 ± 0.92 ^a
Aroma and taste	5.88 ± 1.01 ^b	6.56 ± 0.82 ^a	6.04 ± 0.97 ^{ab}
Juiciness	6.36 ± 0.57 ^{ab}	6.56 ± 0.58 ^a	6 ± 1.11 ^b
Texture	6.56 ± 0.58 ^a	6.44 ± 0.76 ^a	6.28 ± 0.84 ^a
Overall evaluation	6.28 ± 0.61 ^{ab}	6.6 ± 0.5 ^a	6.12 ± 0.78 ^b

Lowercase letters indicate a significant difference between samples (p <0.05)

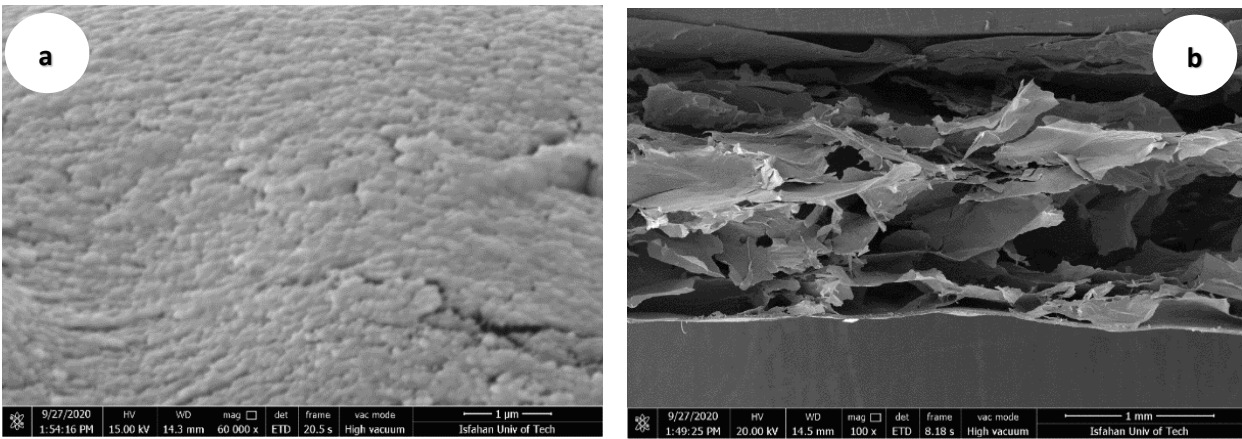
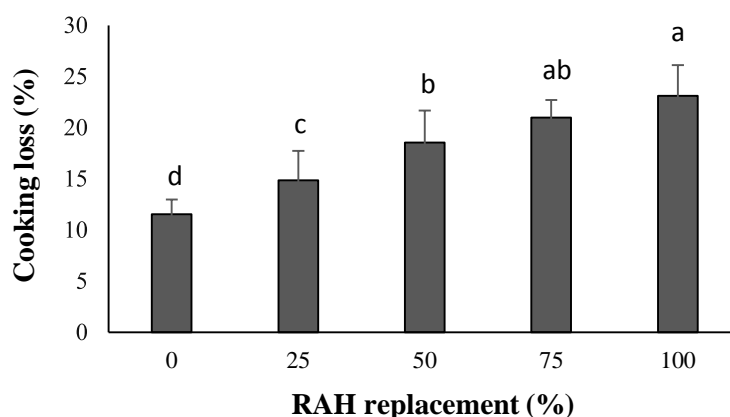


Figure 1. FE-SEM images of agar cryogel from a) surface and b) cross-section.



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517 **Figure 2.** Cooking loss of sausages prepared with different amounts of RAH. Small letters
 518 indicate a significant difference between the samples ($p < 0.05$).
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520 تولید سوسیس فرانکفورتی کم چرب با استفاده از یک جایگزین چربی جدید: هیدروژل آگار بازسازی شده

521 راضیه رنجبران، و هاجر شکرچی زاده

522 روند رو به رشدی به سمت محصولات گوشتی سالم با محتوای چربی کمتر وجود دارد. استفاده از جایگزین های چربی
 523 می تواند مشکلات کیفی محصولات کم چرب را برطرف کند. این مطالعه با هدف تولید سوسیس فرانکفورتی کم چرب با
 524 استفاده از یک جایگزین چربی جدید انجام شد. بنابراین، در مرحله اول، هیدروژل آگار بازسازی شده (RAH) با افزودن آب
 525 به کرایوژل آگار پس از همگن سازی تولید شد. در مرحله دوم، RAH به عنوان جایگزین چربی در سوسیس فرانکفورتی
 526 در غلظت های 0، 25، 50، 75 و 100 درصد استفاده شد و ویژگی های سوسیس کم چرب حاصل از جمله ترکیب شیمیایی،
 527 ظرفیت نگهداری آب، کاهش پخت، ویژگی های بافت، تخلخل، رنگ، درصد چربی پس از سرخ کردن، جذب روغن، pH
 528 در طول زمان و ارزیابی حسی مورد بررسی قرار گرفت. نتایج نشان داد که RAH می تواند دو نوع ژل را با تغییرات دما
 529 تشکیل دهد: یک ژل کم گیر در دمای 55 درجه سانتی گراد و یک ژل با ست شدن بالا در دمای 90 درجه سانتی گراد.
 530 جایگزینی روغن در سوسیس و کالباس باعث افزایش رطوبت، تخلخل، جذب روغن و از دست دادن پخت می شود. از سوی
 531 دیگر، این جایگزینی باعث کاهش محتوای چربی، نیروی برش، ظرفیت نگهداری آب، درصد چربی پس از سرخ کردن و
 532 خواص بافتی مانند سختی، چسبندگی، فنری، صمغی و جویدنی شد. نتایج اندازه گیری pH در طول زمان نشان داد که گروه
 533 های ضد میکروبی سولفات در ساختار آگار، فساد سوسیس های حاوی RAH را نسبت به نمونه شاهد به تاخیر انداخت.
 534 ارزیابی حسی نشان داد که سوسیس های حاوی RAH از نظر رنگ، آبدار بودن و بافت تفاوت معنی داری با نمونه شاهد
 535 ندارند. با این حال، طعم و پذیرش کلی به طور قابل توجهی تحت تأثیر این جایگزینی افزایش یافت ($P < 0/05$). بنابراین
 536 مصرف کنندگان نمونه ی با 25 درصد جایگزینی را به عنوان نمونه بهینه انتخاب کردند. در نتیجه، RAH می تواند با
 537 موفقیت به عنوان جایگزین چربی در محصولات کم چرب با ویژگی های کیفی مطلوب استفاده شود.