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Study of the physicochemical, microbial and sensory properties of hamburgers enriched with turmeric and omega-3 loaded nanoliposomes

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ABSTRACT

In this study, the effect of turmeric extract and omega-3 encapsulated with soy lecithin on the characteristics of hamburger during the 12-day storage period was investigated. The results showed that during storage for 12 days, the highest pH and water holding capacity were related to the control sample. The initial moisture content of hamburgers was about 52%, which decreased significantly after the storage period ($p<0.05$). The lowest shrinkage was compared to the control sample, and the highest cooking loss was observed on the first day of storage. In all the investigated days, the highest volatile nitrogenous compounds was related to the control sample. Free fatty acids increased significantly during storage ($P<0.05$). The samples containing the extracts were not significantly different from the control samples in terms of texture characteristics. Encapsulated treatments had lower bacterial counts than free extracts. The control sample significantly received the lowest score in all sensory characteristics. In general, the quality of hamburgers containing fine-coated turmeric extract and omega-3 improved compared to the control sample.

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1. Introduction

Meat is one of the most important sources of protein, which in addition to protein (essential and non-essential amino acids) contains minerals such as iron and zinc, fatty acids and various vitamins, which make it one of the best and most complete foods [1]. . Today, the consumption of red meat and its products is increasing widely all over the world. One of the most important meat products related to red meat is hamburger, which has attracted the attention of consumers due to its high nutritional value, good taste and easy consumption, as well as the absence of chemical additives in its production process. According to the standard definition, a hamburger made from minced meat of halal animals, especially beef, veal and sheep, with or without added fat and spices and vegetables such as onion and garlic, prepared by a special mechanical device with a weight of 100 grams and frozen in wax paper is offered [2]. Animal meat is a suitable source for the growth of pathogenic microorganisms and parasites and can cause disease in humans through food chains. The growth of microorganisms during the storage period is the main factor in the deterioration of the quality of meat and meat products [3]. The spread of food-related diseases, along with the social and economic problems caused by it, has created the need to produce healthy foods and, accordingly, use new and non-synthetic antimicrobial compounds as much as possible [4]. Since the discovery of chemical drugs and antimicrobial compounds, although it has become possible to control food infections, some bacteria are resistant to the above compounds [5]. In addition, there have been concerns about the safety effect of artificial additives and the side effects of chemical preservatives on human health, which has drawn attention to the discussion of replacing them with natural antimicrobial compounds [6].

Turmeric from the ginger family with a scientific name*Turmeric is long* And it is known by the English name of Turmeric. Curcumin is the active ingredient of turmeric plant. In addition to curcumin, there are many chemical compounds such as volatile oils, alpha and beta tourmarin and other substances such as arabinose, fructose, glucose and starch in the turmeric plant. The color of turmeric is related to color substances such as curcumin, des-methoxy curcumin and bis-demethoxy curcumin [7].

Omega-3 fatty acids are a family of unsaturated fatty acids whose first double bond is located between the third and fourth carbon in the carbon chain [8]. Omega-3 fatty acids are necessary to regulate the activities of the human body, but they are not made in the human body. Omega 3 consists of three fatty acids, which are: alpha-linolenic acid (ALA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Alpha-linolenic acid is found in walnuts, chia seeds, some types of beans, greens, and in soybean oil, canola oil, flaxseed, and olive oil [9]. Two other fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), are found in fish such as salmon, in fish oil, and in fish supplements. ALA, EPA, and DHA are all from the family of n-3 fatty acids (third carbon unsaturated), which are very important from a nutritional point of view because they all have polyunsaturation. The human body does not have the ability to produce n-3 fatty acids (third unsaturated carbon) from other molecules and must obtain it through special foods [10].

Microencapsulation is the process by which compounds are protected from adverse environmental conditions (eg, pH, light, temperature, oxygen, and relative humidity). By doing this, compounds in food or drugs can maintain their performance for a long time [11]. Microencapsulation systems use coating materials such as protein (gluten, whey proteins, isolates, etc.), lipids (glycerides, phospholipids, waxes, etc.) or carbohydrates (starch and their derivatives, cellulose and their derivatives, plant exudates and extracts, etc.). to protect or cover the compound or nuclear material [12]. Microencapsulation of plant extracts and perfumes is increasing due to the important features that this technology creates in these microencapsulated materials, among which it is possible to increase the stability of microencapsulated materials by protecting them from environmental, enzymatic and chemical changes, providing a buffer state. , against pH changes, coping with thermal changes and ionic changes, protection against unpleasant tastes and odors, controlled release of microencapsulated material and complete mixing of immiscible materials [13]. Since the shelf life of products such as hamburgers is of particular importance and according to the contents presented in this research, the purpose

of this study is to investigate the stability of the bioactive compounds of turmeric extract and omega-3 enriched finely coated with soy lecithin on the cooking quality and shelf life of hamburgers during the freezing process and Store in the refrigerator.

2- Materials and methods

2- 1- Materials

Soy lecithin was purchased from Arvin Tejarat (Tehran, Iran). Omega-3 and turmeric were obtained from Sigma (USA) and a local store, respectively. All chemical compounds were purchased from Merck, Germany.

2- 2- Extraction of extract by Soxhlet method

10 grams of turmeric powder was placed in the thimble of the Soxhlet device, and then 120 mL of 96% ethanol was poured into the balloon, and then the balloon was placed in an oil bath under reflux conditions at a temperature of 35 degrees, and the extraction process continued for 18 hours. After the completion of this process, the solvent was separated from the extract by rotary and the concentrated extract was dried in an oven at 35°C [14].

2- 3- micro coating of turmeric extract

For this purpose, in order to prepare 100 cc of nanoliposome from turmeric extract, 0.1 g of the extract after dissolving in 10 cc of 98% ethanol was injected into 100 ml of acetate buffer which was heated to 60 on a magnetic stirrer. The buffer was completely blown. After cooling the solution, lecithin was slowly added to the solution in different ratios of extract to lecithin (1 to 4, 1 to 8, 1 to 12, and 1 to 16) until a completely uniform mixture was obtained after 30 minutes. To form the nanoliposome solution, it was homogenized for 5 minutes with a magnetic homogenizer at 25,000 rpm, then it was subjected to ultrasound with a power of 400 watts for 15 minutes. The prepared liposomes were stored at 4°C for 24 hours before proceeding [15].

Turmeric extract microcoated with soy lecithin was selected at a ratio of 1:12 and the weight ratio of the extract to omega-3 was 1:1. Initially, it was enriched with omega-3 and then, like the steps of microcoating of the extract, this treatment was also microcoated.

4-2- 60% hamburger production

To prepare the burgers, the fresh beef fillet used in this study was prepared from Super Meat located in Sari city, and after three stages of grinding, it was immediately transferred to the laboratory for the production of hamburgers. Other ingredients, including spices, fillers and binders, were added to the meat in the proportions given in Table 1, and all the ingredients were mixed together for 1 hour to obtain a uniform mixture. Then, in order to prepare different treatments, the uniform mixture produced was divided into five equal parts and to each of them (except for the control treatment), the powder of micro-coated extract with the same concentration (obtained in the MIC test) was added, and each treatment was kneaded again until the extract was evenly mixed in the mixture. . Then burgers weighing 100 grams were prepared by a hand mold and placed in zip cap nylons and were tested for 12 days at 4 degrees in the refrigerator to perform physico-chemical and sensory tests on days 0, 4, 8 and 12. .

Treatments: hamburger treatment containing turmeric extract; hamburger treatment containing turmeric extract and omega 3; hamburger treatment containing turmeric nano extract (turmeric extract finely coated with soy lecithin in a ratio of 1 to 12); Hamburger treatment containing turmeric nano extract (turmeric extract finely coated with soy lecithin at a ratio of 1 to 12) enriched with omega 3; Control treatment

Table 1 Hamburger formulation (%)

Meat	Toasted flour	Onion	Whey powder	Wheat flour	Spice	Refined edible salt
60	14.8	14	4.5	3	2.5	1.2

5-2- Tests

2-5-1- Color test

The color of the hamburger was measured using a Colorflex Hunterlab colorimeter (EZ, Virginia, USA). The device was first calibrated with a white tile and then imaging was done. The color test results include three Hunter indices b*, a*, L*, where L* is the symbol of darkness (0) and lightness (100), a* is the symbol of green to red, where -a is green and +a is red, and b* is the symbol It is from blue to yellow, which indicates +b is yellow and -b is blue [16].

2-5-2- pH, moisture percentage and water holding capacity

The pH of the samples was measured by a

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manual pH meter model PP-203 (Taiwan). Moisture percentage was measured by gravimetry under vacuum. 1 gram of the microcapsules obtained from the microcoating process was placed in an oven at 70°C and vacuum once for 6 hours to reach a constant weight:

= Moisture percentage content

100 x coated weight (grams) / moisture weight (grams)

To measure storage capacity[1] water (WHC) of each 5 hamburger samples, 2 grams of samples were placed in two layers of Whatman No. 1 filter paper (weighed) wrapped in a centrifuge tube and centrifuged at a speed of 3000 rpm for 10 minutes at a temperature of 20°C. . After centrifugation, the sample was separated and the filter paper was weighed again [17]. The water holding capacity was performed with 3 repetitions and was calculated and expressed based on the following relationship in terms of 100 grams of water in the sample:

WHC (%) =

100 × Secondary sample weight – primary sample weight

Initial weight of the sample

2-5-3- peroxide index

The amount of peroxide was calculated in milliequivalent grams per kilogram of fatty substance according to the following equation:

= the amount of peroxide in the weight of the oil sample

N * (s-b) 1000 ×

IN

In this relationship, N is the normality of sodium thiosulfate, s is the volume of sodium thiosulfate consumed by the oil sample, b is the volume of sodium thiosulfate consumed by the witness, and w is the weight of the sample [18].

2-5-4- free fatty acids

The amount of free fatty acids in terms of percentage of oleic acid is obtained from the following equation:

(4) ml (0.1 N) NaOH = 0.0282 gr of oleic acid

2-5-5- The amount of shrinkage

The diameter and thickness of the burgers were measured before and after frying and grilling, and their shrinkage was calculated according to the following equation:

= percentage of aggregation

100 × Diameter of cooked hamburger - diameter of raw hamburger

Raw hamburger diameter

2-5-6- The amount of volatile nitrogenous compounds

The amount of volatile nitrogenous compounds in the samples was measured by the method of distillation and Kaldahl titration [17]:

= amount of volatile nitrogen compounds (mg per 100 grams of sample)

4x amount of sulfuric acid consumed (for example)

2-5-7- Measurement of tissue characteristics

All the tested samples were tested with 3 repetitions at room temperature and with texture analyzer (TA-XT2i, Brookfield Engineering Laboratories, Inc., USA). For each treatment, three samples with dimensions of 20x20 mm were separated from the middle of each hamburger and subjected to a two-step pressure test. The samples were compressed up to 40% of their original height with cylindrical balls with a circular cross-section with a diameter of 6.35 mm and a movement speed of 1 mm/s [19].

2- 5- 8- cooking loss

In order to measure the baking efficiency, the weight of each sample was measured before and after baking using a digital scale with an accuracy of 0.001 with 3 repetitions. Baking efficiency was determined using the following relationship:

= percentage of cooking loss

100 × The weight of the cooked product - the weight of the raw product

Raw product weight

2-5-9- Microbial tests

Microbial tests include counting coliforms, mold and yeast and counting*Staphylococcus aureus* They were. Counting of coliforms according to the Iranian national standard method number 9263, counting*Staphylococcus aureus* According to the national standard of Iran No. 6806-1 and mold and yeast counting was done according to the national standard of Iran No. 10899-1 [20].

2- 5- 10- Sensory evaluation

Sensory evaluation was done using a trained evaluator group consisting of 10 people (5 men and 5 women). These people gave their opinions after evaluating the texture, smell, color, taste with a 9-point scale (9 excellent and 1 extremely bad). The critical acceptance point of each of the 5 features was considered, and a

[1]. Water holding capacity

score lower than 5 (average quality) meant rejecting the desired sensory feature [21].

6-2- Statistical analysis

Statistical analysis of samples was done using SPSS version 22 software and data were analyzed by one-way analysis of variance (ANOVA). A completely random design was used and comparison of averages was performed in the form of Duncan's multiple range test at a statistical level of 5%. The tests were performed in 3 repetitions and the graphs were drawn using Excel version 22 software.

3. Results and Discussion

1-3- pH, humidity and water holding capacity

The results of pH changes are shown in Figure 1. The results showed that during storage for 12 days, the highest pH was related to the control sample (hamburger without any additives). On the first day of storage, no significant difference was observed between different treatments, but on other days of storage, microcoated samples had lower pH, which indicated the protective effect of microcoating during hamburger storage. The results were consistent with the Iranian National Standard No. 2304 of hamburger characteristics and showed that the use of fine-coated turmeric extract as an antioxidant can be a suitable alternative to synthetic preservatives in various hamburgers. Similar results were also reported in a study conducted on hamburgers made from pork meat after adding natural antioxidant compounds of grape pomace extract, that the pH did not change after adding these preservatives [22]. The primary meat used to produce hamburger had a pH of about 5.63, which in the control sample increased rapidly to 6.28 during the storage period. The increase in pH in the control sample is due to the accumulation of volatile bases such as ammonia and trimethylamine produced by protein hydrolysis and amino acid decomposition by natural meat enzymes or microorganisms. In the case of the samples containing free and micro-encapsulated extract, the pH increased slightly, but this increase in pH was not significant on different days for each treatment.

Based on the results obtained in Figure 1b, the initial moisture content of hamburgers was about 52%, which decreased significantly after the storage period. After 12 days of storage, the control sample reached a moisture content of 42% and therefore had the lowest moisture content. No significant difference was found between other treatments, which can be attributed to the protective effect of micro-coating and the ability of turmeric and omega-3 bioactive compounds to maintain moisture. Parafati et al. [23] also reported that the microcoated samples lost less moisture in hamburgers containing microencapsulated pear extract.

Figure 1 shows the effect of free and microcoated turmeric extract and omega-3 on the water holding capacity (WHC) of hamburger. The results showed that the highest WHC of the control sample was observed after the storage period. Also, the micro-coated treatments had a higher water holding capacity than the free samples. Microcoating with lecithin was effective on water absorption and viscosity control of hamburger samples, which can be due to the protective role of the wall in preserving nuclear materials during storage. The protective effect of the wall in microcoated products has also been reported by other researchers. Mahdavi et al. reported that by adding barberry anthocyanin extract to the jelly formulation, the water retention of the samples improved [24].

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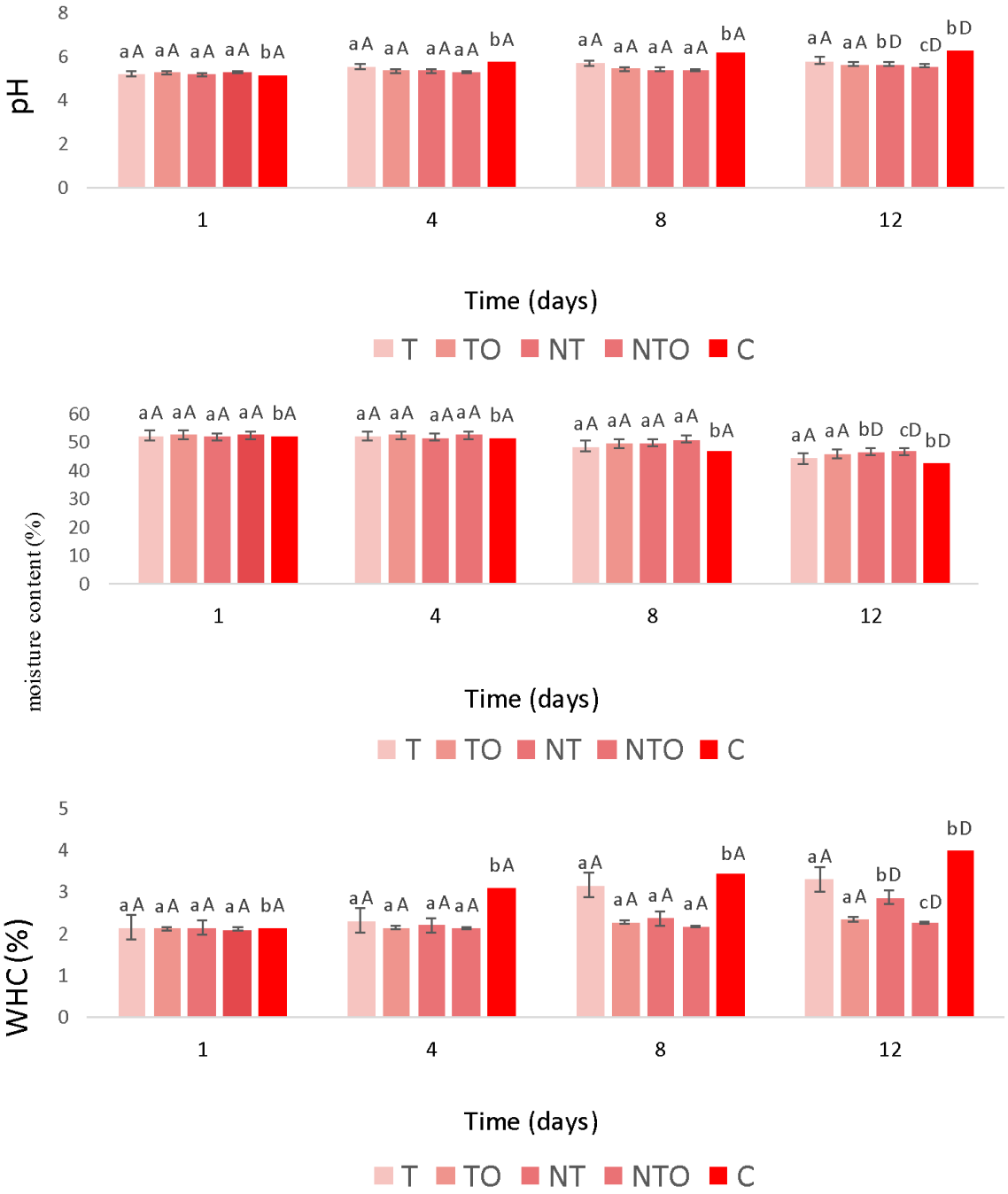

Fig 1 a: pH, b: moisture content and c: water holding capacity of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

2-3- Volatile nitrogen compounds

Changes in the amount of volatile nitrogen bases during 12-day storage at refrigerator temperature can be seen in Figure 2. On the first day, statistically, no significant difference was observed between the values of volatile bases of different hamburger treatments (P>0.05). In all treatments, with the passage of time, from the first day to the twelfth day, the amount of volatile bases gradually increased. In all the investigated days, the highest amount of volatile bases was related to the control sample, adding the extracts to the hamburger samples led to a significant decrease in the amount of volatile bases in the treatments. At the end of the twelfth day, as in other days, the highest amount of TVN was related to the control sample (24.42) and the lowest was obtained in the treatment containing the finely coated extract of turmeric and omega-3 (14.75).

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Various researchers have reported the reduction of volatile bases in the investigation of the effect of adding plant extracts to hamburger. Alaa et al. [25] reported a significant reduction of volatile nitrogenous bases in silver carp fish fillet kept in aqueous extract of bo leaf and rosemary in free and microcoated form. Also, Abdo et al. [26], in the study of nanoemulsions containing curcumin and use in chicken fillets, reported that microcoating led to the reduction of volatile bases during 12-day storage.

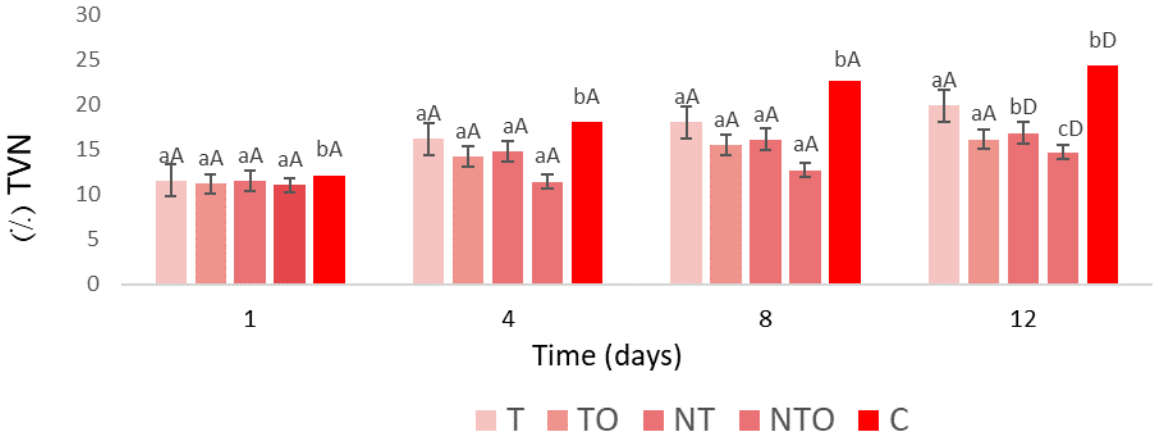

Fig 2 TVN of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

3-3- The degree of hamburger shrinkage

According to the results reported in Figure 3, the rate of shrinkage of hamburgers on the first day of storage was about 17%, which decreased to about 11% after 12 days of storage. After 12 days of storage, the lowest shrinkage was related to the control sample and no significant difference was observed between other treatments.

Shrinkage is an important quality characteristic of meat products that has an important effect on appearance and customer friendliness. In general, the accumulation in the hamburger is related to the absorption and preservation of moisture. Although microcoating leads to moisture retention and less changes in moisture retention, it seems that the addition of turmeric and omega-3 extracts to the formulation has led to a reduction in shrinkage during storage because the protein polymer chain includes lipophilic and hydrophilic groups, in fact, this The polymer chain facilitates the connection of protein with water and fat, as a result, it is able to keep fat and other substances in the form of protein and prevent the extraction of the extract and reduce the shrinkage of the product. Kumar and Kumar [27] did not report significant changes in the treatments with the microcoating of plant extract to meat

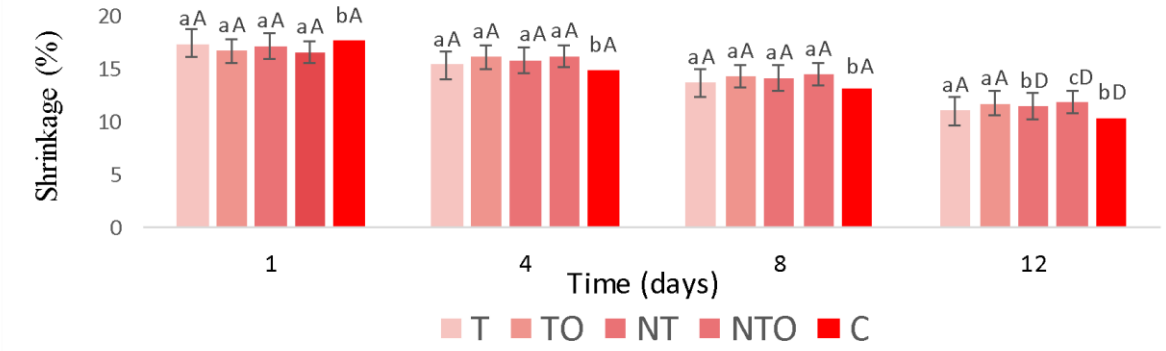

Fig 3 Shrinkage of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

3- 4- cooking loss

Cooking loss is also one of the important criteria of meat products. The main reason for the loss of cooking is the loss of the

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ingredients in the meat due to muscle wrinkling and protein denaturation and a noticeable decrease in water retention capacity. The highest cooking loss was observed on the first day of storage (Figure 4), while after 12 days of storage, the cooking loss of all treatments decreased. The microcoated samples showed less curing loss than the free extracts, which could be related to moisture retention and less shrinkage.

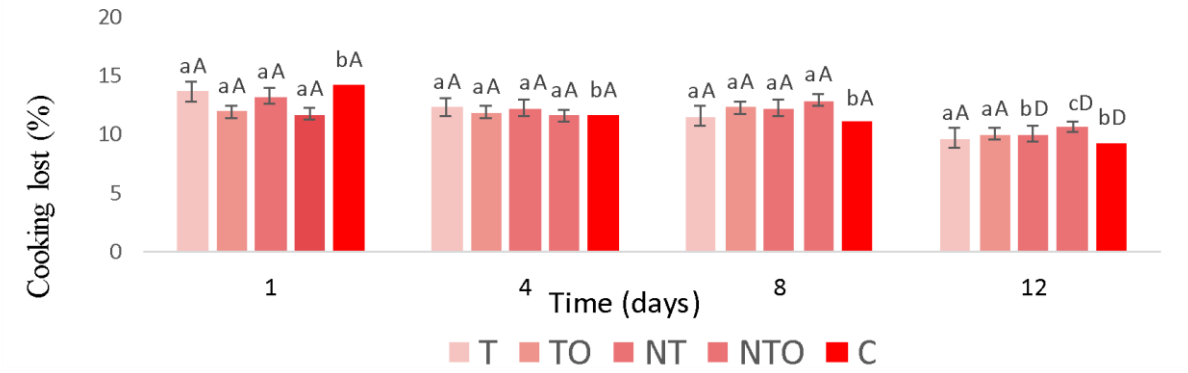

Fig 4 Cooking lost of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

5-3- Free fatty acids

Figure 5 shows the changes of free fatty acids of the samples during 12 days of storage. Based on this, free fatty acids increased significantly during storage, so that the highest free fatty acids were related to the control sample after 12 days of storage. No significant difference was observed between the treatments on the first and fourth days, but on the eighth and twelfth days, microcoated samples had significantly less free fatty acid. The amount of free fatty acid of the samples is related to the oxidation of hamburgers. Due to the protective role of microcoating on nuclear materials, samples containing free extract have more free fatty acid. The samples containing omega-3, despite the high potential of this polyunsaturated fat for oxidation, had less free fatty acid than the control sample due to microcoating. Also in samples containing extractFree turmeric had less free fatty acid than the control sample due to the antioxidant effects of turmeric's bioactive compounds. Mone Kata et al. [28] also reported that the extracts significantly reduced free fatty acids by adding plant extracts with antioxidant properties in a microcoated form to meat.

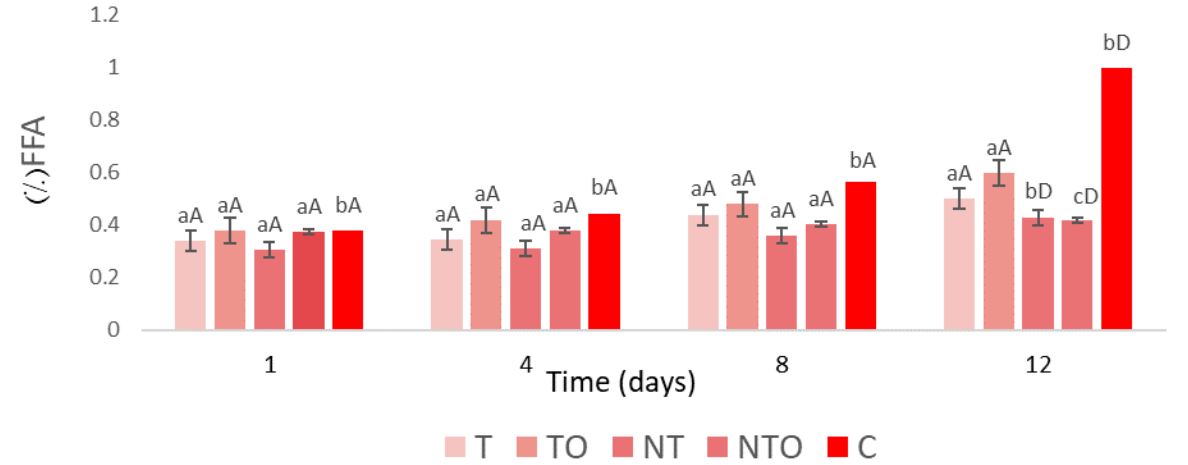

Fig 5 Free Fatty Acids of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

3- 6- number of peroxide

The intensity of oxidation of hamburger containing treatments was measured during 12 days of storage and the results related to peroxide value were reported in Figure 6. The results of analysis of variance showed that the effect of treatment and time on peroxide value was significant ($P<0.05$). Also, the peroxide values of the samples containing different amounts of plant extracts were higher than the control sample on all test days. As can be seen, the control sample had the highest amount of

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peroxide value in all days. All the investigated samples were significantly different from the control sample in terms of peroxide value. The lowest number of peroxide after 12 days of storage was related to the microcoated sample containing turmeric extract. Due to its high oxidation potential, omega-3 led to an increase in peroxide value in samples containing omega-3.

Antioxidants are active for a certain period of time, and with the passage of time, their degree of effectiveness gradually decreases, which can be the reason for keeping the samples in oxidation and heat conditions until they become completely ineffective; Therefore, in the first days, there was no significant difference between the samples containing plant extract and the control sample, but in the last days, as the oxidation reactions increased, the difference between the samples was evident. In fact, the increase in the amount of peroxide can be attributed to the formation of hydroperoxides, i.e. primary oxidation products. The results of Heidarian et al.'s research [29] indicated that the samples treated with rosemary extract were able to reduce the peroxide level of chicken samples in the samples treated with 1% and 3% of the extract to the standard limit of 10 meq/kg in the quality limit. keep desirable and acceptable. In addition, the effect of microencapsulated plant extracts on reducing the peroxide number of meat products has been reported by various researchers [30 and 31].

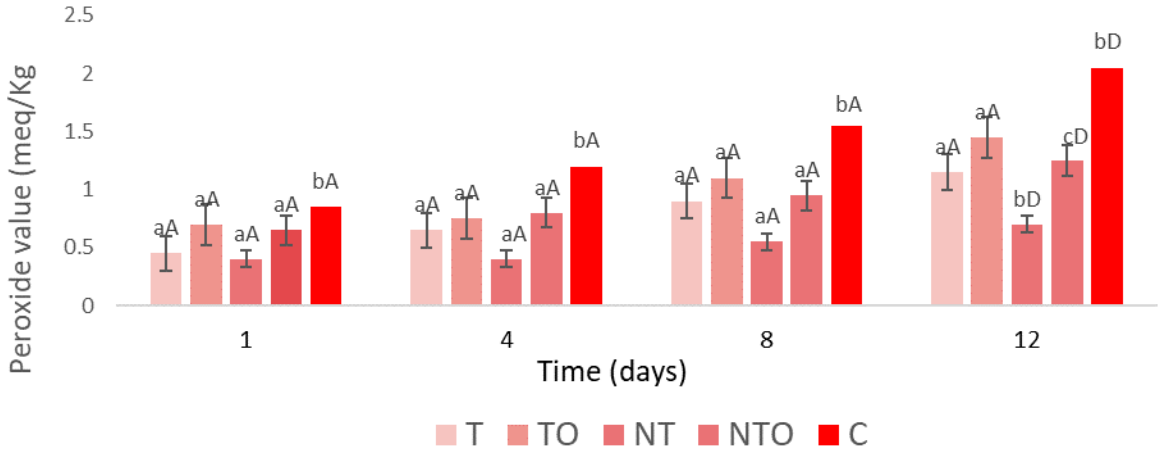

Fig 6 Peroxide Value of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

7-3- Colorimetric factors

The results of the brightness index are reported in the supplementary file. Based on this, no significant difference is observed between different treatments during storage, but the control sample had a significantly lower brightness index during all storage days. The decrease in the brightness of the hamburger samples in the samples during storage is related to the protective effect of the microcoating and also the color of turmeric, which led to the preservation of the brightness. The results of the redness index (a*) showed that the control sample had a significantly lower amount of redness in all days of storage, to the extent that after 12 days of storage, the a index became negative, which means it turned to green. In all days of storage, samples containing microcapsule extract have significantly more red. The results of the yellowness index (b*) showed that the lowest yellowness index was observed in the control sample. The samples containing free turmeric extract had a higher amount of yellowness, which is related to the yellow color of turmeric. Microcoating and use of omega-3 partially reduces the yellowness of turmeric in hamburgers.

8-3- Texture characteristics

Table 2 shows the textural characteristics of the resulting hamburgers. The hardness value of the resulting samples was significantly higher than the control sample. Also, micro-coated treatments had significantly less hardness, which could be due to the effect of micro-coating in maintaining moisture during storage. The resulting coherence range was between 0.69 and 0.78 and the difference was not significant ($P<0.05$). The viscosity trend was similar to the hardness trend. Phenrite in the samples was between 0.81 and 0.93. The

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difference between the treatments in terms of pheneritis was also not significant (P<0.05). The gumminess and chewability had a similar trend.

Table 2 Texture properties of hamburger containing turmeric extract and encapsulated turmeric extract.

Treatment	Hardness (N)	Cohesiveness	Gumminess	Springiness	Chewiness
C	142.33 ± 0.2 a	0.68 ± 0.1 b	96.56 ± 0.1 b	0.92 ± 0.1 a	88.83 ± 0.1 b
T	212.09 ± 0.3 b	0.71 ± 0.3 b	150.58 ± 0.3 c	0.83 ± 0.3 a	124.98 ± 0.3 c
TO	209.85 ± 0.3 b	0.75 ± 0.3 b	157.38 ± 0.3 c	0.88 ± 0.3 a	138.49 ± 0.3 c
NT	218.17 ± 0.5 b	0.73 ± 0.4 b	159.26 ± 0.4 c	0.81 ± 0.4 a	129.00 ± 0.4 c
NOTHING	233.11 ± 0.2 c	0.30 ± 0.1 a	69.93 ± 0.1 a	0.82 ± 0.1 a	57.34 ± 0.1 a

(T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

Different letters indicate significant differences for each parameter for each treatment (p<0.05).

The obtained results confirm the results related to humidity and water holding capacity. In 2014, Haqshana et al. [32] reported that the addition of microencapsulated beta-glucan to the shrimp nugget formulation significantly reduced hardness compared to the control sample.

3-9- Sensory evaluation

In this study, the sensory evaluation of hamburger samples was done by 10 sensory evaluators during the days of storage. The variables investigated in the sensory evaluation included color, smell, taste, texture and overall acceptance, which was assessed by the 5-point hedonic method. A score of 3.5 was set as the threshold for accepting or rejecting the product. In general, the score of evaluators decreased during maintenance. Also, the control sample significantly received the lowest score in all sensory characteristics. No significant difference was observed between other treatments, which indicated the successful addition of free and microcoated extract to the hamburger and the stability of the characteristics during storage.

10-3- Microbial tests

The effect of turmeric and omega-3 extracts on the population*Staphylococcus aureus* It is shown in figure 7. The results of analysis of variance indicate significant effects of treatment type, storage time and their mutual effects. In the control sample, it reached from 1 log CFU/g on the first day to 12 log CFU/g on the twelfth day of storage. In the samples containing free extract, with the passage of time of keeping the population *Staphylococcus aureus* It gradually increased. In hamburgers treated with microencapsulated turmeric extract and omega-3 population *Staphylococcus aureus* reached 6.5 log CFU/g.

Bacterial changes process *Escherichia coli* Also exactly the same as*Staphylococcus aureus* and the lowest amount*Escherichia coli* In all days of storage related to the sample coated with turmeric and omega-3, the highest amount of bacteria was observed in the control sample (Figure 7).

The initial population of mold and yeast in hamburger was 1 log CFU/g, which reached 12 log CFU/g at the end of the storage period. Turmeric essence had an inhibitory effect on the growth of mold and yeast in both free and micro-encapsulated states, but these effects were especially significant in the case of micro-encapsulated essence after 12 days of storage (Figure 7). On the first day of storage, except for the control sample, the other treatments had zero mold and yeast counts, which is due to the antimicrobial effect of the extract and omega-3, which was proven in the previous sections. On other storage days, the lowest amount was related to the microcoated samples and the highest amount was related to the control sample. The free extracts had more yeast and mold than the microcoated samples, which was due to the protective effect of microcoating on the core compounds.

Staphylococcus aureus It is one of the pathogenic bacteria whose presence in meat products will be associated with a serious outbreak of poisoning. Although in the broth dilution test, the difference between MIC and MBC of free and microencapsulated turmeric extract for*Staphylococcus aureus* It was not observed, but in the food system, different results regarding the effect of the micro-covering process on population reduction*Staphylococcus aureus* seen. The reason for this difference can be attributed to the presence of different strains of*Staphylococcus aureus* In the natural microbial population attributed to meat.

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Generally *Staphylococcus aureus* Due to being gram positive, it is more sensitive to plant extracts or their compounds. According to Jonosite et al. [33], the addition of herbal flavorings in minced meat stored at refrigerator temperature causes population reduction. *Staphylococcus aureus* became. Fingerroot microencapsulation was carried out in two-layer emulsions stabilized with chitosan-lecithin and its antimicrobial effect on pork spoilage index microorganism was investigated within 15 days [34]. The results showed that the studied perfume in both free and micro-covered states had a significant effect on population reduction.*Staphylococcus aureus*and among the investigated bacterial populations (total microbial count, lactic acid bacteria and total forms),*Staphylococcus aureus* It was the most sensitive bacteria to perfume. According to the national standard of Iran (2014), population*Staphylococcus aureus*Coagulase positive in frozen or fresh minced meat samples should not be more than 10 CFU/g^3×5 (equivalent to 6.69 log CFU/g). The results of this research show that at the end of the storage period, only the samples containing turmeric extract and microencapsulated omega-3 are within the permissible limits of the Iranian standard. One of the health indicators for evaluating meat and meat products is the presence of Enterobacteriaceae bacteria. In this research, the average initial count of bacteria*Escherichia coli* in the flesh

The first day was zero, which indicates the application of proper sanitary conditions during the preparation of hamburgers. Since Gram-negative bacteria are generally considered to be resistant to natural antimicrobial compounds due to the specific characteristics of their outer membrane, nevertheless, the beneficial effects of adding turmeric extract and omega-3 in a microencapsulated state in reducing its population were very noticeable. The results of the microbial count showed that the addition of free and microcoated turmeric extract in the fresh hamburger formulation reduced the growth of microbial populations.*Escherichia coli* Wow*Staphylococcus* It was found that these effects can be related to the antimicrobial properties of turmeric extract and omega-3. Micro-coated treatments had less bacteria than free extracts, which is due to the protective effects of micro-coating and increasing the shelf life of extracts by protecting them against decomposition, oxidation and evaporation [35]. In addition, the stable release of extracts from nanoparticles during storage provides the possibility of maintaining the antimicrobial effects of extracts [36]. Due to the fact that in food systems like buffer environments, it is not possible to completely release the extract loaded in nanoparticles, therefore micro-encapsulated perfumes exerted their antimicrobial effects on the investigated populations in lower concentrations [37]. The antifungal effects of turmeric essence are related to the destruction of cell membrane structure and vesicles, disturbance in the biosynthesis of ergosterol regulating membrane fluidity and changes in the structure of cells during proliferation. Also, disturbance in Ca balance^{2+} and H+ in the cell, disruption of gene expression, calcium-related stress, disruption of ergosterol synthesis, disintegration of cell membrane, and nutritional starvation are among the most important mechanisms related to the antifungal effects of turmeric extract and curcumin [38, 39]. Regarding the confirmation of the increase in antifungal activity of plant essences after microcoating, various researchers have reported this effect, which is consistent with the results of the current research on increasing the inhibitory effects of turmeric extract and omega-3 on mold and yeast after microcoating.

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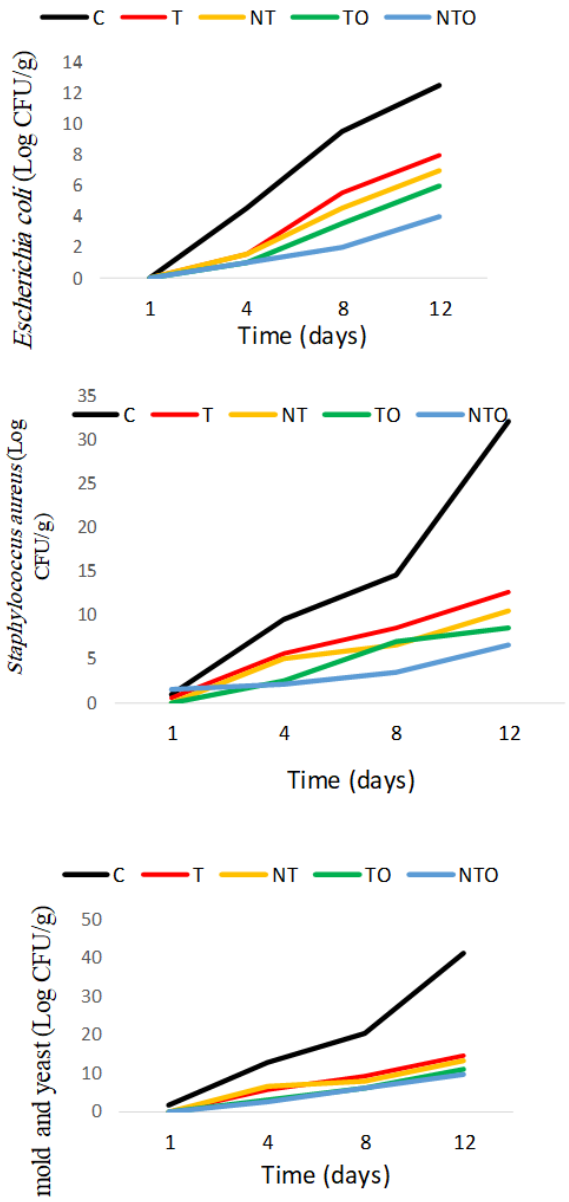

Fig 7 *Escherichia coli*, *Staphylococcus aureus* and mold and yeast count of hamburger containing encapsulated extract during 12 days storage at 4 °C (T: hamburger containing turmeric extract, TO: hamburger containing turmeric extract and omega3, NT: hamburger containing encapsulated turmeric extract, NTO: hamburger containing encapsulated turmeric extract and omega3, C: Control).

5- General conclusion

In this study, the effect of turmeric extract and omega-3 microcoated with soy lecithin on the characteristics of hamburger during the 12-day storage period was investigated. During storage for 12 days, the highest pH was related to the control sample, i.e. hamburger without any additives. On the first day of storage, no significant difference was observed between different treatments, but on the other days of storage, microcoated samples showed lower pH, which indicates the protective effect of microcoating during hamburger storage. The initial moisture content of hamburgers was about 52%, which decreased significantly after the storage period. After 12 days of storage, the control sample reached a moisture content of 42% and therefore had the lowest moisture content. The results showed that the highest water retention capacity was related to the control sample after the storage period. Also, the micro-coated treatments had a higher water holding capacity than the free samples. The results of hamburger textural characteristics showed that the samples containing the extracts did not differ significantly from the control samples in terms of textural characteristics. Micro-coated treatments had less number of bacteria than free extracts, which is due to the protective effects of micro-coating and increasing the shelf life of extracts by protecting them against decomposition, oxidation and evaporation. In addition, the stable release of the extract from nanoparticles during storage provides the possibility of maintaining the antimicrobial effects of the extracts. Sensory evaluation showed that the evaluators gave a high score to the treated samples and in their opinion there was no significant difference between the control sample and the treated samples in terms of sensory. In general, the results showed that the microencapsulated extracts could be successfully used in the hamburger formulation, so that the characteristics of this product were improved.

6- Resources

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تأثیر عصاره زردچوبه و امگا۳ ریزپوشانی شده با لسیتین سویا بر خصوصیات فیزیکوشیمیایی، میکروبی و حسی همبرگر

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چکیده

در این مطالعه تأثیر عصاره زردچوبه و امگا۳ ریزپوشانی شده با لسیتین سویا بر خصوصیات همبرگر طی دوره ۱۲ روزه نگهداری بررسی شد. نتایج نشان داد در طی نگهداری به مدت ۱۲ روز، بیشترین pH و ظرفیت نگهداری آب مربوط به نمونه شاهد بود. میزان رطوبت اولیه همبرگرها، حدود ۵۲ درصد بود که پس از دوره نگهداری کاهش معنی‌داری یافت ($P<0.05$). کمترین جمع شدگی مربوط به نمونه شاهد بود و بیشترین افت پخت در روز اول نگهداری مشاهده شد. در همه روزهای مورد بررسی، بیشترین میزان باز فرار مربوط به نمونه شاهد بود. اسیدهای چرب آزاد طی نگهداری افزایش معنی‌داری یافت ($P<0.05$). نمونه‌های حاوی عصاره‌ها ازلحاظ ویژگی‌های بافتی تفاوت معناداری با نمونه‌های شاهد نداشتند. تیمارهای ریزپوشانی شده تعداد باکتری کمتری نسبت به عصاره‌های آزاد داشتند. نمونه شاهد به‌طور معنی‌داری کمترین امتیاز در کلیه خصوصیات حسی را دریافت کرد. به‌طورکلی کیفیت همبرگرهای حاوی عصاره ریزپوشانی شده زردچوبه و امگا۳ نسبت به نمونه شاهد بهبود یافت.