Development Of Sausages Fortified With Flaxseed Oil Microcapsules And Its Impact On Oxidative And Sensory Stability

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Abstract

Background: Meat is considered beneficial to health due to its nutritional composition but lack some essential nutrients except meat from marine sources like omega-3. These fatty acids perform a variety in functions in normal development of body, but their use is limited in functional foods due to their oxidation. Freshwater fishes are good source of protein, but fat content of their body does not fulfill the daily omega-3 requirements.

Methods: Current study was aimed to fortify freshwater fish sausages with omega-3 fats from flaxseed oil. Flaxseed oil was microencapsulated using chitosan and the powdered microcapsules were added in sausages batter at four different concentrations i-e 2.5%, 5%, 7.5% and 10% in T1, T2, T3 and T4 respectively. sausages also prepared without fortification of microcapsules and kept as control. Direct addition of flaxseed oil (bulk flaxseed oil, BFO) was done for comparison. Proximate composition, color, texture, water holding capacity, cooking loss, fat loss, lipid oxidation and sensory analysis was performed at different storage intervals.

Results: Proximate composition of sausages did not show any significant change except for fat content. texture, color, water holding capacity, fat loss, cook loss were significantly influenced in treatment and control batches ($p \leq 0.05$). lipid oxidation values were between the acceptable ranges in all batches except sausages prepared with BFO have high values of CDs (0.79 µmol/mg sample) and TBARS (1.13 mg MDA/Kg sample).

Conclusion: Sensory evaluation of sausages was satisfactory at different storage intervals and T1, T2 and T3 have shown highest overall acceptability values and these levels can be used to prepared enriched sausages with flaxseed microcapsules without any side effect and acceptable oxidative stability.

Keywords: Flaxseed oil, Fortification, Microencapsulation, Omega-3, Sausages,

INTRODUCTION

Our diet small part constitutes lipid, but its contribution is high in terms of flavor, taste, texture, palatability, color and most important is source of energy. Omega-3 is group of polyunsaturated fatty acids are known as essential fatty acids due to their crucial role in human body, including Alpha Linolenic acid (ALA), Eicosapentaenoic acid (EPA), Decosahexaenoic acid (DHA) Omega-3 fatty acids mainly obtained from marine fatty fish and vegetable oils like flaxseed. These fatty acids should be incorporated in diet due to normal growth and development of body as

human are unable to produce omega-3 in their bodies. Omega-3 fats play vital role in controlling cardiovascular diseases, inflammation, hypertension, cancer and enhance immunity (Bazan, 2018; Bahadoriet al. 2010; Caugheyet al. 2010). Market working with omega-3 related products have shown elevated trend with a business of USD 2.04 billion during year 2016 and still growing at a rate of 6.6% from 2012-2020. Interestingly it is estimated that asia pacific grow at 7.5% from year 2015-2022 (Grand View Research Report, 2018). Consumption of omega-3 fats among human is not according to recommended intake of official agencies (Rubio-Rodríguez et al. 2010). To achieve daily dietary intake of recommended intake, 2 serving size fishes must be consumed by a person in a week which is not fulfilled resulting in deficiency of omega-3 fats (Kolanowski and Weißbrodt, 2007). Fresh water fishes have great nutritional value, but they have low content of omega-3 fats due to their feeding habits (Li et al. 2011). Human body needs proteins and omega-3 fats too. Highest ratio of omega-3 to omega-6 among plant sources is found in flaxseed oil (National Research Council, 1993). Its inclusion in meat products is advantageous to enhance nutritional value and palatability is also improved as compared to marine fish oils (Wang et al. 2016).

ALA is major component of flaxseed oil with values ranging from 57-60% as compared to other fatty acids. Metabolism of ALA in human body resulted in high content of EPA and DHA in blood plasma after ingestion of flaxseed oil (Valenzuela et al. 2014). Deficiency in consumption of ALA consequently diminishes the formation of its end products EPA and DHA. Both EPA and DHA are main part of membranes as phospholipids, helps in normal development of brain and retina (Simopoulos, 2002). Due to excessive use of saturated fats consumption, high rate of cardiovascular diseases is reported (O'Flaherty et al. 2012). There is interest to develop diets with low amount of saturated fats. People have awareness regarding sources of diet containing low fat is healthier while those having saturated fats, exert bad impact on health (Bolger et al. 2017; Diekman and Malcolm, 2009). Additionally, deficiency of omega-3 fats induced due to high ratio of linoleic acid containing diets that hinders the conversion of ALA into EPA and DHA and contributed to deficiency of omega-3. Diets containing sunflower oil, corn oil, peanut should be avoided as they have high content and linoleic acid which contributed in lowering omega-3 fatty acids (Naeem et al. 2019). It is needed to enhance the dietary uptake of omega-3 containing oils like flaxseed oil (Zahran and Tawfeuk, 2019; Ramcharitaret al. 2005).

There is an increase in utilization and demand of functional foods fortified with omega-oils. Meat based products are consumed 3-4 time in a week showing their high consumption. Sausages are meat products that can be fortified with omega-3 enriched oil from plant sources like flaxseed oil (Asuming -Bediakoet al. 2014; Pelseret al. 2007). Many reviews have been published on health benefits of flaxseed oil (Kajlaet al. 2015; Goyal et al. 2014). It was reported by European Food Safety authority that 0.3g/100g content of ALA in food products is helpful in lowering blood cholesterol levels (EC, 2012). However, incorporation of flaxseed oil in products is limited due to high rates of susceptibility to oxidation that consequently, damage and affect products shelf life during storage period (Asuming-Bediakoet al. 2014; Berasategiet al. 2011). Many techniques have been used by researcher like use of antioxidants to preserve essential oils (Yesilsu and Ozyurt, 2019; Ankabiet al. 2018). Another approach to protect the bioactive omega-3 oils is microencapsulation. It is contemporary technique to preserve the hydrophobic materials from oxidation, temperature and light during storage (klinkesorn and McClements, 2009). Oxidative deterioration of plant oils like flaxseed, grapeseed and walnut oil was secured due to formation of covering over the hydrophobic core material that act as a physical barrier. Encapsulated oils were produced in the form of powders that masks odor of oils, render their regulated

release and enable handling during use (Kaushik et al. 2015; Encinaet al. 2016).
Taking account of all the aspects, this study was aimed to develop fish sausages fortified with different level of flaxseed oil microcapsules to enhance their nutritional profile and content of omega-3 fatty acids and to assess the physical and oxidative properties of sausages at different storage intervals.

Materials and Methods

Biological material

Rohu fish was obtained from UVAS fisheries research farm, degutted, filleted and stored at -20C till their further use in preparation of sausages. Flax seeds were purchased from local market and their oil is extracted using mini oil presser machine. Chitosan and Maltodextrin were procured by Sigma chemicals Lahore, Pakistan.

Reagents

Food grade glacial acetic acid was used for emulsion preparation while methanol, sulfuric acid, chloroform solutions were employed for proximate composition as reagents. Methanol, chloroform, isopropanol solution was used in Fatty acid profile analysis. 2-propanol, perchloric acid, thiobarbituric acid and malonaldehyde bis 97% was used for evaluation of oxidative stability of fish sausages.

Experimental plan

Fish sausages were prepared with four different concentration 2.5% (T1), 5% (T2), 7.5% (T3) and 10% (T4) of microcapsules of flaxseed oil. A control batch of fish sausages was also prepared without fortification of microcapsules. Flaxseed oil was extracted using mini oil presser machine. Fish sausages were vacuum packed and stored at -4^{o}C and randomly selected for chemical and textural analysis after their formation, 7 days, 14 days.

Flaxseed oil microencapsulation

Chitosan and Maltodextrin was dissolved in water prior to emulsion formation. Chitosan solution (1.5% optimized in another study) was prepared in 0.50ml/100ml of acetic acid solution. It was constantly stirred until complete mixing of contents. Wall material solutions (MD & CS) were kept on shaking water bath overnight in order to hydrate polymer molecules and to obtain dissolution of material. Core wall material with ratio 1:4 was added in formulations and subjected to laboratory homogenizer operated at 5000 rpm for 30 minutes for emulsion formation. Emulsions after preparation were subjected to a laboratory scale spray dryer (Toption Lab Spray Dryer, Xi'an, China) machine with a nozzle diameter of 1.5mm, inlet temperature 180 ^{0}C, outlet temperature 80 ^{0}C and 6ml/min emulsion pumping rate. Emulsion was constantly stirred before being fed to spray dryer machine to avoid amalgamation of oil droplets. Powdered microcapsules were stored in airtight glass container soon after their formation. Flaxseed oil microcapsules obtained retaining single cyclone separator, packaged, and stored at -4^{o}C temperature until their addition in fish sausages. Experiments were executed in triplicates.

Sausage formation

Fish sausages were prepared according to the method described by Dincer and cakli (2010). Fish fillets were used for preparation of sausages following their mince formation using kitchen food processor. Ingredients were added and final mixing done with addition of flaxseed oil microcapsule powder. Better was poured in casing and covered with PVC film and refrigerated and analyzed at bdifferent storage intervals. Different batches of sausages were prepared as depicted in table. A control batch of sausages was made without addition of microencapsulated powder while in formulation 1, bulk flaxseed oil was added. Formulation 2, 3 ,4, 5 were obtained through addition of 2.5%, 5%, 7.5% and 10% flaxseed oil microcapsules, respectively.

Proximate analysis

Moisture, protein, fat and ash content of fish sausages were determined according to AOAC (1995) method.

Water holding capacity.

Expressible moisture designated as water holding capacity (WHC) evaluated using method described by Zhuang et al. (2007). Briefly, 10g sample from each batch uncooked sausage was separately mixed with 0.5M NaCl (15ml) solution and transferred to a centrifuge tube, vortexed for 1 minute followed by storage in refrigerator for 15 minutes. Resulting contents were centrifuged at 3000 rpm for 15 minutes. Analysis was performed in triplicates. WHC was calculated according to formula:

$WHC\% = (W_{pellet} - W_{uncooked}/W_{uncooked})$

W_{pellet}denotes the solid formed at bottom after centrifugation.

$W_{uncooked}$designatethe initial weight of sausage used for analysis.

Water and fat binding capacity

Schuh et al. (2013) method was used for measurement of water and fat binding capacity of sausages. Uncooked sample (24g) from each batch was transferred to 50ml capacity centrifuge tube and placed on heating water bath temperature set at 98C for 45 minutes. Tubes were placed on ice bath for 10 minutes for cooling of contents. After cooling of mixture, tubes were inverted on a pre-weighted porcelain dish for an hour and released fat was noted by weighting the dish again. Total fluid loss was evaluated by difference between weight of porcelain dish after release of fat and before inversion and expressed as total fluid loss (%). Water loss (WL) was measured by placing the dish having released fluid in an oven at 100C for 24 hours. WL (%) was calculated by difference before and after drying sample. Finally fat loss (FL) was obtained by subtraction of WL from TFL.

Cooking loss

Cooking loss was determined according to the method described by Bolger et al. (2017) after 3 days of sausage refrigerated storage and expressed as initial weights percentage. Cook look was calculated using formula:

Cook loss (%) = [Uncooked sausage weight (g) – cooked sausage weight(g)]/ uncooked sausage weight (g)× 100 (Sikes et al. 2009)

Texture analysis

Sausages were kept at room temperature for 30 minutes before texture analysis using texture profile analyzer. Sausages from each batch were cut into 2cm cylindrical portions with a diameter of 2.5cm. a cylindrical probe of 2.5 com was used to compress the samples 75% of their original height twice at a cross head speed of 0.80mm/s. Mechanical properties of sausages (hardness, chewiness, gumminess, springiness) were evaluated from the formation of curves as result of compression (Savadkoohiet al. 2014).

Color

Instrumental color of fish sausages in all batches was measured using tintometer by placing a transparent film on both sides of sample for the determination of (L*) luminosity, (a*) redness and (b*) yellowness. Tintometer was calibrated on white tile before its use over samples (Solomando et al. 2020).

Sensory analysis

Samples sensory analysis was performed using a 6-point hedonic scale for unpleasant to extremely liked inscriptions during different storage intervals (first day, 7 days, 15 day) of storage. A penal of 20 untrained volunteers evaluated sensory attributes of samples including color, texture, aroma, appearance, flavor and overall acceptability. Each panelist was served with fragments of sausages on white plastic plates from each batch and provided with glass of water (Kairamet al. 2021).

Induction period

Rancimat technique (Professional Rancimat, 892) was employed for determination of induction period. Samples (2.5g) from each treatment were subjected to dry oxygen at 120 °C for a period of one hour. Induction period at different storage intervals was expressed through break point of graph curves at 0 day, 7 days, 15 days and after frying (Ullah et al. 2020).

Thiobarbituric Acid Reactive Substances

TBA test was applied on products to measure the secondary oxidation of PUFAs because

this test is easy to perform and precise. 1ml of encapsulated oil was be added in a test tube containing 2 ml of thiobarbituric acid reagent. Test tube were shaken well, vertex and were kept on boiling water bath for 15 min and after cooling centrifuged at 1000rpm for 15 min. Supernatant obtained was used for absorbance in spectrophotometer (Hitachi U-2000, Tokyo, Japan) at 532nm against blank. TBARS was expressed as mg of malonaldehyde/Kg sample (McDonald and Hultin 1987).

Conjugated dienes (CDs)

Evaluation of CDs was performed according to procedure labeled by Juntachote et al. (2005). Frozen fish nuggets were thawed at room temperature and minced using mincer followed by addition of distilled water (5mL) and formation of slurry using homogenizer. Extraction solution(5mL) of 3:1 (v/v) hexane/isopropanol was poured in 0.5ml of slurry then it was subjected to centrifugation at 5000rpm for 5 min for separation of supernatant. Absorbance of extracted supernatant was taken at 233nm using spectrophotometer and resulted were expressed as micromoles per sample Kg.

Statistical analysis

Experiment was carried out with three replications and each analysis was performed thrice. Data was analyzed using statistical software SAS 9.1 and means difference was considered significant at $p \leq 0.05$ and evaluated through ANOVA and DMR test was used to test the significance.

Results

Chemical composition of sausages was determined through proximate analysis. Table 1 showed the proximate composition of sausages at first day prepared with microcapsule inclusion at different concentration. It was evident from results that fortification did not impact the protein content of sausages from different treatments groups and control. Fat content showed significant difference amount different formulations. Highest fat content was observed among T4 having 10% microencapsulated flaxseed oil supplementation. Highest moisture content was recorded for T4 while lowest values were noted for sausages from control batch that were made without microcapsules enrichment. There was a decreasing trend in moisture content of sausages with increase in supplementation. Ash content ranged between 3.28-2.40% in studied batches.

Color

Table 6.1 showed the changes in color parameters of fish sausages from different formulations. Color of products is considered as key parameter for sausages as consumers are inclined towards bright and characteristic color of sausages. Lightness (L^*) of sausages showed a decreasing trend with increasing fortification of flaxseed oil microcapsules. Sausages prepared with direct addition of flaxseed oil have bottommost values of lightness while control sausages value recorded were highest among treatment sausages. Yellowness (a^*) of sausages showed an increasing trend with an increase in addition of flaxseed oil microcapsules. Control batch peak values were noted for BFO and smallest for control groups.

Water holding capacity

WHC is an important attribute related to sausages ability to hold moisture. Water holding capacity was higher in T3 as compared to other treatments. Values were significantly different among treatments having different level of microcapsule inclusion and control. Lowest moisture content was observed in sausages having direct addition of flaxseed oil. It indicated that microcapsule addition being added change the water holding capacity. Microcapsules were prepared using chitosan and maltodextrin. These ingredients effectively increase the water holding capacity of sausages.

Fat and cooking loss

Fat loss evaluated for different batches of sausages and recorded in table 2. Fat loss values ranged between 1.22-0.70 for different studied treatments. Highest values of fat were observed for control experiment sausages while lowest were recorded for sausages

prepared with 7.5% flaxseed oil microencapsulated powders. There was a significant difference noted in different treatments except T2 and T4. Little difference among values of fat clearly indicated low loss of water from cooked sausages during cooking. After cooking if sausages, cooking loss values were significantly higher in T4 samples as compared to control and bulk flaxseed oil addition in sausages. Difference in cooking loss of sausages with various levels of fortification can be explained on the basis of moisture content values. Lowest moisture content in T4 (10%) consequently has more dry matter due to powder, thereby mostly affected by cooking loss.

Texture analysis

Table 6.2 illustrated that textural analysis of sausages with different enrichment levels, there was significant difference among hardness of sausages from different batches. Highest values of springiness and gumminess were recorded for formulation control batches. Values of springiness were ranged between 0.95-0.970 with control sausages showed highest springiness and T3 formulation the least one. There was a changing trend observed for different formulation textural properties with the level of inclusion and in control samples. Decreased cohesiveness in control and more hardeness in control were attributed to oil addition in treated groups. Springiness and gumminess were increased with decrease level of fortification and high in control sausages.

Lipid oxidation

Figure 2 represented the conjugated dienes (CDs) and Thiobarbituric acid reactive substances values in fish sausages enriched with various levels of flaxseed oil microcapsules and in control and BFO groups. CDs values were not influenced by the enrichment level after processing of sausages and no significant difference was observed between control and treatment batches. CD s value recorded for Control, BFO, T1, T2, T3 and T4 were 0.11, 0.12, 0.10, 0.11,0.12 µmol/mg of sample respectively. At second storage interval values fluctuate and there was significant difference noticed between control and treated groups. CDs in control group after 7 days was 0.14 µmol/mg of sample while for BFO was 0.22 µmol/mg of sample. Highest values of CDs were observed for T4 and lowest for T1 and T3. Evaluation of CDs at third storage interval showed an increasing order except for T3 whose readings were lowest among the enriched batches. Sausages having direct addition if flaxseed oil (BFO) were shown to have highest CDs values after 14 days of storage. At 4th storage interval effect of cooking was evaluated in control and modified sausages. There was an increase observed in CDs after cooking in all batches of sausages. Cooking did not have strong impact on oxidation in T1 and T2 with CDs 0.35 and 0.33 µmol/mg sample respectively and difference was not significant.

TBARS values assessment at different storage intervals was presented in table. Raw sausages after their preparation showed no significant different in values of TBARS and values for control, BFO, T1, T2, T3, T4 were 0.16, 0.16, 0.15, 0.18, 0.16, 0.18 mg MDA/Kg of sample, respectively. Influence of storage on TBARS after 7 days period in different raw baches of sausages showed an increasing trend with highest TBARS values noticed for BFO sample sausages and lowest in T2 and T4. TBARS values were evaluated after 14 days storage interval and a significant difference was observed between control and treated groups. Elevated values of TBARS in BFO as compared to other treatments means microencapsulation protected the bioactive component of sausages. Control sample increase in TBARS values was lower as compared to samples enriched with 10% microcapsules of flaxseed oil. Cooking effect was also analyzed after 14 days of storage. Raw sausages and cooked sausages values of TBARS were influenced in different batches. Lower values of TBARS were observed after cooking for control group sausages while highest values were recorded for BFO sample. Control batch sausages values were lower as compared to T4 batch and high as compared to T1, T2, T3 batches. Cooked sausages values ranged between 0.37- 1.13 mg MDA/Kg.

Sensory Evaluation

Results of sensory analysis were analyzed at different storage interval after cooking at different storage intervals. A 9-point hedonic scale was used for sensory evaluation of sausages. Omega-3 enrichment levels in fish sausages significantly impact the sensory attributes. Lowest scores for color were observed for BFO batches after a storage period of 14 days. Sensory attributes of fish sausages having direct addition of flaxseed oil were largely affected as compared to control and treatment batches. At 0 days interval sausages flavor values were not fluctuated in treatments and control sausages showed best scores, on the other hand BFO revealed lower. Flavor scores were not different when evaluated after 7 days and slight change in treatment flavor values observed after 14 days with highest and lowest scores noted for control and BFO sample, respectively. High values for texture were found for T1, T3 and T1 after 0 days, 7 days and 14 days. Scores of overall acceptability were satisfactory for fortified batches and control batches when observed at different storage periods. On the contrary lowest values of overall acceptability were recorded in BFO batches results of sensory attributes were related to oxidative stability analysis of control and treated samples.

Discussion

Our findings are in accordance with previous studies on moisture content of enriched products. Studies conducted by Aquilani et al. (2018) on cooked pork burgers having fish oil microcapsules supplementation and they observed low moisture content in control batches as compared to treated samples. Protein content was not significantly different which means supplementation did not change the proximate composition (Josquin et al. 2012). However, fat content was changed due to microcapsule addition in studied groups. WHC is a key factor related to meat products acceptability among consumers. It is related to loss of water during processing storage, cooking and it affects the texture and meat juiciness (Warner et al. 2017). Results agree with previous findings by kucukcetin et al. (2011) while working with gelatin and gum Arabic as stabilizers in yogurt. They found an increase in WHC values proportionately with stabilizer addition. Similar results were obtained by Pancaret al. (2016). Heck et al. (2017) working with pork burgers enriched with vegetable oil microcapsules and observed higher values in treated and control samples. Another study by Josquin et al. (2012) found lowest moisture content in treatments groups having microencapsulated oil that contributed to cooking loss. Similar behavior in cooking loss was also reported by Bilek and Turhan (2009). Samples with lower levels of fat resulted in lower cooking loss ($p \leq 0.05$) and vice versa. Lower cooking loss is helpful in retention of fat level in products which can be explained based on high thermal stability of fortified products. Fat retention and cooking loss are important attributes of meat-based products that affect their meat matrix and sensory qualities (Serdaroglu, 2006; Aaslynget al. 2003). Textural change is a challenge for the development of good quality meat products. Results are important in describing the addition of plant-based microcapsule addition in fish-based meat products. Sausages prepared with microcapsules addition were harder as compared to direct addition of flaxseed oil sausages (Pires et al. 2020). Similar investigations were made by Josquin et al. (2012) and Pelser et al. (2007). They found that inclusion of microencapsulated fish and flaxseed oil contributed to enhancing firmness of sausages. Different levels of inclusion of flaxseed oil microcapsules had a different impact on textural properties of sausages (Shi et al. 2014). Increased vegetable oil addition in meat products changed the network formation and physicochemical properties and which in turn tenderness of meat products.

Current study result of product color was different from findings of Solomondo et al. (2020) that checked the impact of monolayered and multilayered fish oil microcapsules addition in chicken sausages and observed no change in color of sausages upon addition. Results of sausages instrumental color were related to finding of

Wu et al. (2021). Color properties of chicken surimi fortified with various levels of flaxseed oil were significantly influenced. An increase in redness (b^*) values was observed as compared to control as reported by Ozvural et al. (2016).

CDs and TBARS tests were performed for evaluation of levels of lipid oxidation products in fish sausages in control and treated batches. Scientific investigations are in not in bulk for CDs assessment in products. Flaxseed oil fortified products shown to have oxidation due to contact with oxidation, storage conditions and temperature. Solomondo et al. (2020) evaluated the impact of single and multilayer fish oil microcapsule addition in chicken sausages and found similar result for CDs. Increase in lipid oxidation of sausages was also observed during storage (Josquin et al. 2012). There was a correlation between freshness of fish products and values of TBARS. Fresh meat products TBARS value should fall below t 0.58 mg MDA/Kg of sample while products having vales of TBARS ranged between 0.58-1.51 mg MDA/Kg of sample were considered as acceptable but have slight rancid. Samples of fish tissues having values higher that 1.51 mg MDA/Kg sample were professed as rancid. Results of current study revealed that TBARS values in T1, T2, T3 were perceived as good and within acceptable range. Sausages prepared with higher content of flaxseed oil microencapsulated powder have values higher than 0.58 mg MDA/Kg of sample and therefore perceived as slightly rancid. Based of our results, 2.5%, 5% and 7.5% levels of enrichment at -4oC storage were acceptable. Current study results were in agreement with previous investigations of lipid oxidation of fortified nuggets, Pork burgers and Spanish salchichon with microcapsules (Lorenzo et al. 2016; Aquilianiet al. 2018; Jimenez et al. 2016).
Current study finding of sensory analysis were similar investigations described by Valencia et al. (2006a) and Martinez et al. (2012) who pointed out changes in flavor and fishy smell due to addition of direct oil in products. However, studies conducted by Aquiliani et al. (2018) on burgers enrichment with microcapsules and found changes in sensory attributes. Keenan et al. (2015) described the presence of fishy smell and off flavors in burgers fortified with omega-3 oil and reported high acceptability values for control groups.

Conclusion

Freshwater fish sausages enriched with flaxseed oil microcapsules were developed first time to enhance content of omega-3 fatty acids. Fortification of flaxseed microcapsules at 2.5%, 5%, 7.5% and 10% significantly changed the omega-3 content of freshwater sausages. Fish sausages prepared FROM T1, T2, T3 batches were shown higher oxidative stability and acceptable sensory values. It is concluded that these levels can be used to enhance the omega-3 content of sausages of fish.

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Table 1: Proximate composition and color scores of fish sausages having different levels of flaxseed microcapsules enrichment

Formulations	Parameters						
	Protein content	Fat content	Moisture content	Ash content	L*	a*	b*
Control	14.24±0.02[a]	13.60±0.22[e]	69.08±0.53[a]	2.40±0.12[c]	68.87±0.04[a]	1.24±0.01[d]	13.69±0.04[f]
BFO	13.16±0.03[a]	14.16±0.01[d]	68.32±0.01[a]	2.47±0.01[c]	63.40±0.14[f]	1.46±0.01[c]	18.40±0.18[a]
T1	11.27±3.36[a]	14.85±0.03[c]	65.21±0.01[b]	2.43±0.01[c]	67.19±0.04[b]	1.24±0.01[d]	14.50±0.15[e]
T2	14.70±0.02[a]	14.92±0.04[c]	64.77±0.52[b]	2.74±0.03[b]	65.93±0.01[c]	1.53±0.04[b]	14.94±0.02[d]
T3	14.52±0.02[a]	15.90±0.23[b]	62.91±0.46[c]	3.11±0.05[a]	65.53±0.19[d]	1.63±0.02[a]	15.41±0.04[c]
T4	13.36±0.20[a]	17.03±0.31[a]	60.82±0.31[d]	3.28±0.02[a]	64.17±0.03[e]	1.65±0.08[a]	16.14±0.08[b]

Means within a column from a parameter having different letters were considered statistically significant at p≤0.05.
Samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4).

Table 2: Texture profile of fortified fish sausages in control and treatment batches.

Formulations	Parameters				
	Hardness (N)	Springiness (ratio)	Cohesiveness (mm)	Gumminess (N)	Chewiness
Control	1.04±0.01[b]	0.95±0.01[a]	0.43±0.02[d]	0.81±0.05[a]	0.85±0.04[a]
BFO	0.92±0.01[c]	0.82±0.02[b]	0.61±0.05[a]	0.68±0.01[b]	0.66±0.01[b]
T1	0.98±0.01[c]	0.78±0.02[c]	0.51±0.01[c]	0.55±0.01[c]	0.44±0.01[c]
T2	1.51±0.02[a]	0.73±0.02[d]	0.59±0.01[b]	0.44±0.02[d]	0.38±0.02[d]
T3	1.63±0.01[d]	0.70±0.02[e]	0.62±0.01[a]	0.41±0.01[d]	0.32±0.01[e]
T4	1.50±0.03[a]	0.76±0.04[c]	0.65±0.01[a]	0.31±0.01[e]	0.23±0.02[f]

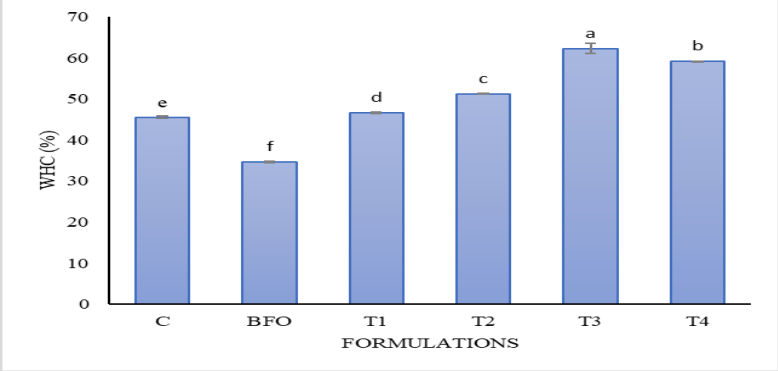
Means within a column from a parameter having different letters were considered statistically significant at p≤0.05.
Samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4)

Table 3: Oxidative stability of sausages prepared with various levels of microencapsulate flaxseed oil powders at different storage intervals.

Formulations	Storage days	CDs (µmol/mg sample)	TBARs (mg MDA/Kg sample)
Control	0 days	0.11±0.01[i]	0.16±0.01[o]
	7 days	0.14±0.01[h]	0.24±0.01[m]
	14 days	0.16±0.01[g]	0.58±0.01[e]
	After cooking	0.22±0.01[f]	0.61±0.01[d]
BFO	0 days	0.12±0.01[i]	0.16±0.01[o]
	7 days	0.22±0.01[f]	0.91±0.01[c]
	14 days	0.56±0.01[d]	1.03±0.06[b]
	After cooking	0.79±0.00[a]	1.13±0.01[a]
T1	0 days	0.10±0.01[i]	0.15±0.01[o]
	7 days	0.15±0.01[g]	0.28±0.01[m]
	14 days	0.23±0.02[f]	0.31±0.02[i]
	After cooking	0.35±0.01[e]	0.37±0.00[k]
T2	0 days	0.11±0.01[i]	0.18±0.01[o]
	7 days	0.18±0.02[g]	0.25±0.01[m]
	14 days	0.23±0.01[f]	0.41±0.01[i]
	After frying	0.33±0.02[e]	0.47±0.01[g]
T3	0 days	0.12±0.01[i]	0.16±0.01[o]
	7 days	0.15±0.92[g]	0.26±0.01[m]
	14 days	0.21±0.01[f]	0.37±0.01[k]
	After cooking	0.54±0.03[d]	0.51±0.01[g]
T4	0 days	0.11±0.03[i]	0.18±0.02[o]
	7 days	0.19±0.02[g]	0.25±0.01[m]
	14 days	0.34±0.01[e]	0.60±0.01[f]
	After cooing	0.57±0.01[d]	0.81±0.01[d]

Means within a column presented with different letters were statistically significant.
Samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4).

Figure 1: Water holding capacity of sausages prepared with different concentration of flaxseed microcapsules



Samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4).

Figure 2: Evaluation of total fat loss (TFL) in flaxseed microcapsule enriched sausages.

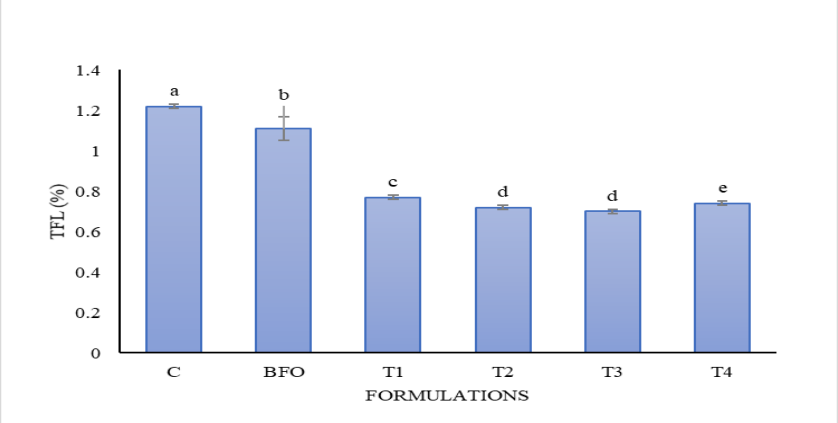
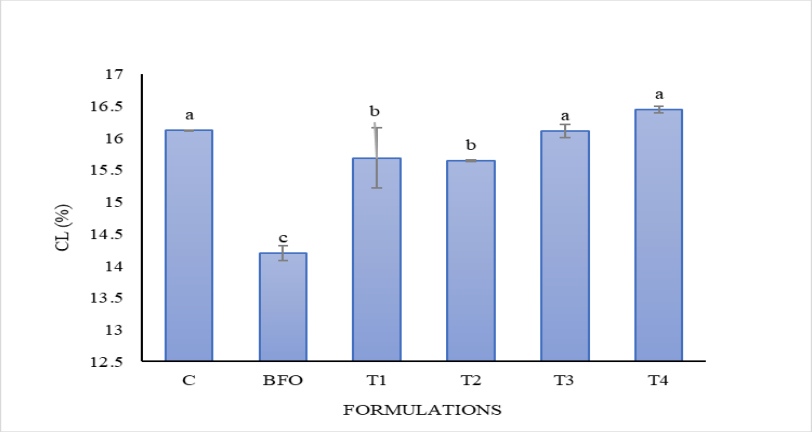


Figure 3: Results on cooking loss of sausages from different batches



samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4).

Figure 4: Sensory analysis of fish sausages enriched with different levels of flaxseed oil microcapsule at 0 day

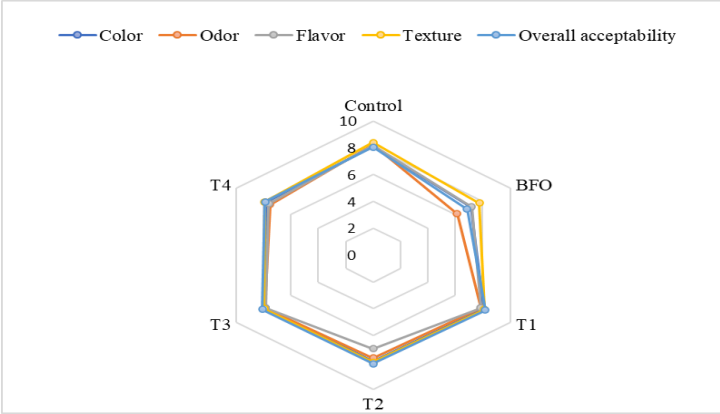


Figure 5: Sensory analysis of fish sausages enriched with different levels of flaxseed oil microcapsule After 7 days

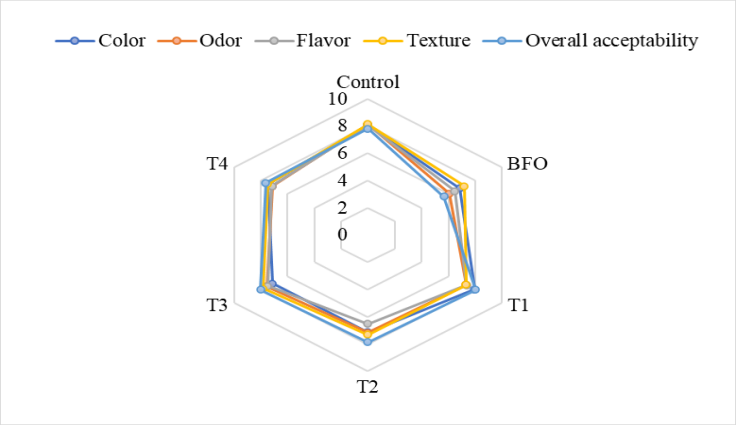
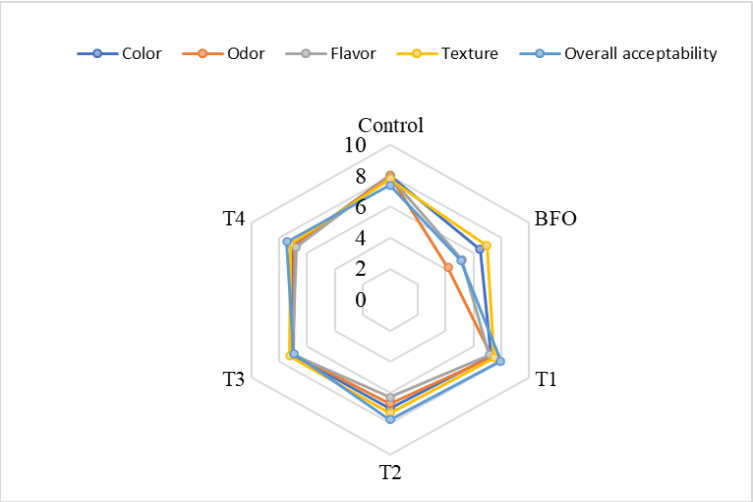


Figure 6: Sensory analysis of fish sausages enriched with different levels of flaxseed oil microcapsule after 14 days.



Samples without fortification of flaxseed oil (C), samples with addition of bulk flaxseed oil (BFO), Samples with 2.5% enrichment of flaxseed microcapsules (T1), Samples with 5% enrichment of flaxseed microcapsules (T2), Samples with 7.5% enrichment of flaxseed microcapsules (T3), Samples with 10% enrichment of flaxseed microcapsules (T4).