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Enhancing functional properties and lipid profile of *Grammoplites suppositus* (Troschel, 1840) sausages through chia seed fortification

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ABSTRACT

Spotfin flathead, *Grammoplites suppositus*, an underutilised marine fish, was utilised for sausage preparation. The proximate composition of the fish - moisture, protein, fat and ash content was determined as 80.71 ± 0.96, 16.29 ± 0.55, 0.56 ± 0.21 and 1.15 ± 0.04, respectively. Fish meat was incorporated at 70% to develop the sausages, which were then standardised through sensory evaluation and labelled as control sausages (CS). The developed sausages were further fortified with chia seeds at 2%, 4% and 6% (w/w) respectively and denoted as CFS samples. The proximate composition (moisture, crude protein, crude fat, total ash, crude fibre, carbohydrate), functional properties (pH, water-holding capacity, cooking loss, total expressible fluid, folding test), physical properties (colour, texture), organoleptic characteristics (colour, odour, texture, flavour, overall acceptability) and fatty acid profile were analysed. A significant difference was observed between all the parameters in the CS and CFS samples. However, among the fortified samples, no significant difference was observed in the pH and b* values, while heterogeneous values were noticed for water holding capacity, cooking loss, emulsion stability, L* and a* values ($p<0.05$). Although CFS 6% samples exhibited better functional properties, the colour and texture values were not sensory-acceptable. Consequently, the CFS 4% samples recorded the highest overall acceptability score. Further, the polyunsaturated fatty acid (PUFA) content of sausages was enhanced through chia seed fortification.

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1. Introduction

The Platycephalids, commonly known as flatheads, constitute the major fishery resource of East Asia, Australia, the Indo-West Pacific, the Eastern Mediterranean, the Red Sea, East Africa and Japan. They hold significant commercial value due to their nutritional potential, flavour, and therapeutic properties (Cheng *et al*., 2019). According to Fairbridge (1960), flatheads were the first fish targeted by the Australian commercial trawl fishery, commanding a high price due to their premium meat quality. *Grammoplites suppositus*, commonly known as the spot-fin flathead, is a Platycephalid that is commonly harvested in the Arabian Gulf, Bay of Bengal, and Sri Lanka but is underutilized in certain other parts of the world (Batool and Siddiqui, 2020). They are listed under the sustainable category of the southwest trawl fishery according to the marine fish stock status of the Central Marine Fisheries Research Institute (2023). Being a prevalent species among the demersal bycatch exploited by the multiday fleet, they have been reported to constitute 2.11 % of the total catch in Karnataka (Abdurahiman *et al*., 2007). Despite having good meat quality and yield percentage, their underutilization may result from the lack of awareness, which minimises their significance in comparison to other economically valued fish.

However, to ensure sustainable consumption and limit the over-exploitation of commercially valued stock, the lesser-known species should be promoted maximally by developing restructured products (Silva *et al*., 2020). The proximate composition studies of the deep-sea scorpaeniformes validate their potential for use in food applications to maximize their economic value (Nihal *et al*., 2023). Sausage is a concentrated meat form blended with additives, enclosed in casings, and then heat-processed to achieve the desired shape and texture. Primarily prepared from traditional meat sources such as chicken, beef, and pork, they often contain high levels of cholesterol, nitrites, sodium, and saturated fats, leading consumers to shift their preferences to seafood products. Fish, an easily digestible protein with high biological value compared to any other form of meat, is well-known to enhance global nutritional security by preventing malnutrition in developed and underdeveloped countries (Silva *et al*., 2020; Nair, 2005) and promoting zero hunger based on the United Nations Sustainable Development Goals (SDG 2). Fish can be an alternative protein source to traditional meat, especially for sausage production (Cardoso *et al*., 2008). Studies regarding fish sausages prepared from the trawling-mix category (Guimaraes *et al*., 2018) and croaker (Cortez-Vega *et al*., 2013) have reported excellent nutritional benefits.

Several authors have reported the benefits of including dietary fibre in sausage formulations (Gore *et al*., 2022; Salazar *et al*., 2021; Pires *et al*., 2020). The addition of dietary fibre to seafood products could improve their functional properties such as texture, gelling ability and water-binding (Borderias *et al*., 2005). Dietary oat fibre added to carp sausages improved gelling characteristics (Gore *et al*., 2022), pea and chicory root fibre incorporated in the formulation of low-fat fish sausages exhibited good textural properties (Cardoso *et al*., 2008), and banana flour added to frankfurter sausages reported an improvement in its nutritional stability (Salazar *et al*., 2021). Chia seeds (*Salvia hispanica*) are superfoods that constitute about 30-34 % of dietary fibre content, 18-24% protein, and the

powerhouse of exogenous amino acids, which are used as foam stabilisers, suspending agents, and emulsifiers for food and pharmaceutical purposes (Kulczynski *et al*., 2019). Moreover, they are gluten-free, possess water-holding ability, and have a strong potential for gel-forming (Arifin *et al*., 2021). Chia seeds are the richest source of omega-3 and omega-6 fatty acids such as alpha-linolenic and alpha-linoleic acids respectively, and can be used as a source of antioxidants for commercial purposes (Ali *et al*., 2012). Chia seeds added to meat sausages have reported several benefits including the prevention of lipid oxidation in pork sausages (Scapin *et al*., 2015), improvement of the sensory and nutritional content of chicken sausages (Arifin *et al*., 2021), and enhancement of the lipid profile of bologna-type sausages (Pires *et al*., 2020). Studies regarding the utilisation of *G. suppositus* in value-added product development and the inclusion of chia seeds into fish sausage formulations have not yet been explored. Hence, this research is designed to standardise sausages from *G. suppositus* and fortify them using chia seeds to improve their nutritional, functional and sensory characteristics.

2. Materials and Methods

2.1 Collection of raw materials

The spotfin flathead (*G.suppositus)* was collected from a multi-day trawler from the Munambam harbour, Cochin, India (10.1667° N 76.1833° E). The average length and weight of the fish were 28.8 ± 0.12 cm and 178.56 ± 0.24 g, respectively. The fish were headed, gutted and filleted manually in batches maintaining a temperature of 2 - 4 ºC. The fillets were packed in polythene bags, labelled and stored in an air-blast freezer (Labline instruments, Cochin) at -18 ºC until sausage production. Whole chia seeds (*Salvia hispanica*) were ground into a fine powder and kept in an air-tight container. All the ingredients for sausage production were sourced from a local supermarket in dry powdered form. Cellulose casings of 22 mm were used for sausage preparation.

2.2 Preparation and fortification of sausages

Sausages were prepared using the recipe of Chattopadhyay et al. (2023) with slight modifications. Four formulations were prepared- control sausage (CS) and chia-fortified sausages (CFS 2%, CFS 4% and CFS 6%). The chilled mince was ground with 1.5% (w/w) NaCl, 0.25 % STPP and 5.25% fat, followed by the addition of potato starch (8% w/w) as the binder, along with the ingredients outlined in Table 1. Chia seeds were added at 2 %, 4 % and 6% respectively to the sausage batter. The comminuted batter was stuffed into cellulose casings (22 mm diameter) using a manual sausage stuffer of 5L capacity (Sun Labz Equipments, Chennai), and linked into a uniform length of 12 cm. It was kept for one hour in the refrigerator (4 ºC) to infuse the flavours into the meat. The resultant sausages were subjected to cooking at a temperature of 80 – 90 ºC for 1 hour in a boiling water bath to an internal temperature of 72 ºC, measured using a digital thermometer. The sausages were then cooled in ice (1:1 w/w) at a temperature of 2 – 4 ºC for 15 minutes and then reboiled in the water bath at 100 ºC for one minute to remove any wrinkles present over the sausages. The sausages were vacuum-packed (Sevana, QS 500, India) in 12-micron polyester and 75-micron Low-Density Polythene (LDPE) pouches and kept in the refrigerator (4 ºC) until further analysis.

2.3 Determination of proximate composition

The proximate composition of *G.suppositus* and the sausage samples was estimated according to AOAC (2005). Moisture content was determined using the AOAC method 945.15, crude protein using the 945.18 Kjeldahl method, and crude fat and total ash using the AOAC method 927.43 and 963.15 respectively. The crude fibre was estimated according to AOAC 978.10 and the carbohydrate content was calculated by deducting the sum of moisture, crude protein, crude fat, total ash, and crude fibre from 100%.

2.4 Determination of functional properties

The pH of the samples was measured according to Carballo *et al*. (1995) using a digital pH meter (Eutech Instruments, Singapore). The water-holding capacity (WHC) was analysed according to Verbeken *et al*. (2005). The cooking loss and the total expressible fluid (TEF) were assessed according to Choi *et al*. (2010) and Hughes *et al*. (1997), respectively. The folding test was conducted using the method given by Lanier (1992).

2.5 Determination of physical properties

The colour parameters, including the L* (Lightness), a* (red-greenness) and b* (yellow-blueness) values were recorded in triplicate using a colourimeter (Hunter Lab Miniscan ® XE plus, USA). The texture profile analysis was done using a texture analyser (Lloyd Instruments, UK).

2.6 Sensory analysis

Sensory analysis was done by a panel of 30 members using a nine-point hedonic scale. The parameters included colour, odour, texture, flavour and overall acceptability.

2.7 Determination of fatty acid profile

The fatty acid profile of the sausages was analysed according to Metcalfe *et al*. (1966). Out of the three CFS formulations, the sausage with superior sensory scores was taken for analysis along with the CS samples. The lipid fraction of the samples in the form of fatty acid methyl

Table 1. Standardised sausage recipe from *G. suppositus* and substitution using chia seeds

Ingredients (%)	CS	CFS 2%	CFS 4%	CFS 6%
Fish mince	70	68	66	64
Chia seeds	-	2	4	6
Salt	1.5	1.5	1.5	1.5
Sugar	1	1	1	1
Sodium tripolyphosphate	0.25	0.25	0.25	0.25
Pepper powder	0.5	0.5	0.5	0.5
Garlic powder	0.5	0.5	0.5	0.5
Onion powder	0.5	0.5	0.5	0.5
Ginger powder	0.5	0.5	0.5	0.5
Potato starch	8	8	8	8
Ice/ Ice water	12	12	12	12
Sunflower oil	5.25	5.25	5.25	5.25

*To ensure consistency in the moisture-to-solid ratio across formulations, the reduction in fish mince was compensated by the addition of ice during mixing

esters (FAME) was analysed using gas chromatography (Thermo Scientific TR-Frame, Australia; Column length: 120 m; film thickness: 0.25 µm; 0.25 mm diameter)

2.8 Statistical analysis

The results are expressed as mean ± standard deviation and analysed using Analysis of variance (ANOVA) to find out the significant difference ($p<0.05$) between the samples using the IBM SPSS program v.25 for Windows. Duncan's multiple range test was used to compare the mean value of the formulations. The graphs were plotted using RStudio 4.3.2.

3. Results and Discussion

3.1 Proximate composition of *G. suppositus*

The proximate composition of *G. suppositus* is shown in Fig. 1. A comparatively higher moisture content (80.71 %) was observed for spotfin flathead, similar to that reported by Venugopal and Shahidi (2012) for haddock, pollock, silver hake, Atlantic halibut and cod. The crude protein percentage in *G. suppositus* was higher than that of *Nemipterus japonicus* and comparable to the values reported by Mohanty *et al*. (2019) for other selected food fishes. More than 15% of protein in fish muscle suggests that it can provide between 30% and 50% of the daily value percentage of protein required by the human diet (Mohanty *et al*., 2019). The crude fat content of the fish recorded lower values, indicating that *G. suppositus* is a semi-fat fish with 0.56 % lipid content. Regarding the total ash content, *G.suppositus* recorded a value of 1.15 %, consistent with the findings of Mohanty *et al*. (2019) for other food fishes. The proximate results were in agreement with Nihal *et al*. (2023), who reported that *G. suppositus* is a low-oil high-protein fish with further utilization potential for developing nutritionally enriched food products, beyond utilizing it for producing fish feed and fertiliser.

3.2 Proximate composition of sausages

The proximate composition of the sausage samples is given in the Table 2. The CS samples exhibited a moisture content of 68.79 % compared to 56.46 % for the CFS 6 % samples ($p<0.05$). This reduction can be attributed to the addition of chia seed in dry powder form, which decreased the moisture content of the product. According to Munoz *et al*. (2012), the efficiency of chia seeds in absorbing 12 times their mass in water further reduces the expressible water content of the sausage substrate. A similar reduction in moisture content was reported by Fernanadez-Lopez *et al*. (2019) for frankfurters with chia seeds added as whole seeds, flour, and co-product. A significant increase in protein content was observed in CFS 6% samples compared to all other formulations ($p<0.05$) (Table 2). Despite the substitution of fish protein with chia seeds, the protein content of the CFS sausages showed higher values. This might be due to the high content of proteins and essential amino acids in chia seeds, possessing high thermal stability and emulsifying capacity (Grancieri *et al*., 2019). Since sausages are essentially emulsion-based products, the network development during gelation is significantly influenced by the functioning of the proteins involved (Elavarasan *et al*., 2024). The myofibrillar protein content in the fish muscle combined with chia seed fortification, contributed to the textural improvement in fortified sausages, similar to the observations made by Elavarasan *et al*. (2024) in millet-fortified sausages. The results align with Pintado *et al.* (2016), who observed an increased protein value for chia-added frankfurters. A significant increase in the crude fat content was notable in the CFS samples, with a 5-fold increase in fat content for the CFS 6 % samples ($p<0.05$). According to Ores *et al.* (2018), the fat content of sausages positively impacts their sensory characteristics, contributing to aroma and flavour development. The results highlight the application of chia seeds as a healthy fat substitute by enriching the food with polyunsaturated fatty acids and preserving its quality attributes (Kulczynski *et al*., 2019). A similar increase in crude fat was observed by Ding *et al*. (2018) for the chia-substituted restructured ham-like products. The ash content of the samples also recorded a similar increase for the CFS samples, as chia seeds contain minerals like phosphorus, calcium, magnesium and potassium in higher amounts (Kulczynski *et al*., 2019) compared to CS samples. The crude fibre content of the CFS samples also showed an excellent increase compared to the CS samples with CFS 6% showing the highest value ($p<0.05$). A similar increase

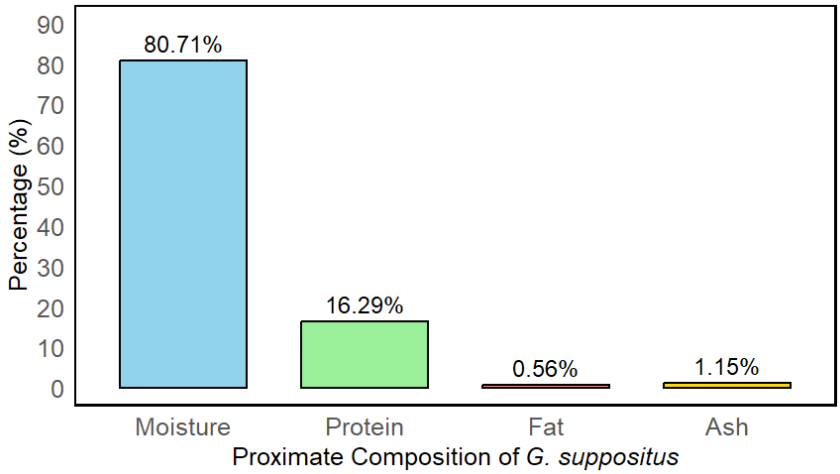


Fig. 1. Proximate composition (%) of *G. suppositus* expressed as mean ± standard deviation (n=3)

Table 2. Proximate composition, colour, texture profile, and folding test scores of the sausage formulations

Parameter	CS	CFS 2%	CFS 4%	CFS 6%
Moisture	68.79 ± 0.67^{c}	62.47 ± 0.20^{b}	59.84 ± 2.49^{b}	56.46 ± 1.19^{a}
Protein	19.98 ± 0.62^{b}	20.15 ± 0.43^{b}	19.70 ± 0.24^{b}	17.97 ± 0.24^{a}
Fat	0.35 ± 0.04^{a}	2.16 ± 0.15^{b}	3.50 ± 0.20^{c}	5.11 ± 0.14^{d}
Ash	0.23 ± 0.02^{a}	1.16 ± 0.07^{b}	2.20 ± 0.07^{c}	2.86 ± 0.33^{d}
Carbohydrate	10.23 ± 0.02^{a}	13.24 ± 0.95^{b}	13.85 ± 0.09^{c}	16.30 ± 0.40^{d}
Fibre	0.42 ± 0.03^{a}	0.82 ± 0.02^{b}	0.87 ± 0.03^{b}	0.97 ± 0.01^{c}
L*	57.83 ± 0.60^{d}	46.44 ± 0.96^{c}	43.52 ± 1.03^{b}	40.49 ± 0.30^{a}
a*	1.71 ± 0.05^{a}	2.59 ± 0.04^{b}	2.93 ± 0.05^{c}	3.25 ± 0.08^{d}
b*	14.48 ± 0.17^{c}	12.69 ± 0.05^{b}	12.36 ± 0.16^{b}	9.77 ± 0.64^{a}
Hardness (N)	26.31 ± 2.58^{c}	23.02 ± 0.42 ^{b}	19.98 ± 0.38^{a}	18.17 ± 0.66^{a}
Cohesiveness	0.45 ± 0.06^{c}	0.44 ± 0.05^{bc}	0.43 ± 0.06^{ab}	0.43 ± 0.05^{a}
Springiness (mm)	7.67 ± 0.20^{c}	7.61 ± 0.11^{c}	6.86 ± 0.31^{b}	6.35 ± 0.02^{a}
Chewiness (Nmm)	66.93 ± 3.81^{ab}	63.98 ± 1.62^{a}	62.29 ± 2.77^{a}	69.71 ± 2.66^{b}
Gumminess (N)	11.36 ± 0.73^{c}	11.46 ± 0.84^{c}	8.83 ± 0.11^{b}	7.07 ± 0.72^{a}
Folding test	3.33 ± 0.57^{a}	3.66 ± 0.57^{ab}	4.33 ± 0.57^{bc}	5 ± 0.00^{c}

in crude fibre content was reported by Attalla *et al*. (2017) for yoghurt mousse fortified with chia seeds. Together with the higher protein composition, the increase in fibre, lipid and ash content, lowered the percentage of carbohydrates in the CFS samples by computing their percentage by difference. The CFS 6% samples showed a significantly lower amount of carbohydrate content (9.28%) compared to the CS samples (12.24%) (Table 2).

3.3 Functional properties of sausages

3.3.1 pH values

With the increase in chia seed concentration, there was a proportionate decrease in the pH level of the samples (Fig. 2a). The CS samples recorded the highest pH value (6.42) followed by CFS 2% (6.33), CFS 4% (6.27) and CFS 6% (6.21) samples. Although a significant difference existed between the pH values of CS samples and all the chia-fortified samples, the pH of CFS 4% and CFS 6% samples exhibited homologous values ($p>0.05$). The reduction in pH values of the CFS samples can be linked to the substitution of fish meat with chia seeds, which reduced the fish meat's natural buffering capacity. However, the high amount of protein and fibre present in chia seeds can interact with various acids and bases in the food matrix, creating a buffering effect (Nakov *et al*., 2024). In addition, the presence of polyphenols like chlorogenic, gallic, caffeic, ferulic and cinnamic acids, epicatechin, rutin, kaempferol, apigenin, quercetin and p-coumaric acid (Kulczynski *et al*., 2019) in chia seeds might have contributed to the lowering of the pH of the sausage matrix. The decrease in pH is beneficial for the retardation of spoilage bacteria and can provide characteristic colour and flavour to the product (Ores *et al.,* 2018). Similar results were obtained by Alvarez *et al.* (2020), who stated that chia seeds can be used as an alternative to synthetic acidulants to reduce the pH values of meat products naturally.

3.3.2 Water Holding Capacity (WHC)

The WHC of sausages is depicted in Fig. 2b. During food processing, drip loss is a major challenge, that leads to product shrinkage, and reduction in sensory acceptance, impacting the tenderness and juiciness of the product (Cheng and Sun, 2008). However, in the study, with chia seed powder addition, the WHC of the CFS samples significantly improved than the CS samples (50.19 %) with the highest value shown by the CFS 6 % samples (70.87 %) ($p<0.05$). The soluble fibres found in chia seeds, primarily glucuronic acid and neutral sugars, produce hydrophilic mucilage that binds water and lowers the viscosity of the substrate (Melo, 2015). This improves the WHC while lowering free moisture levels by strengthening the sausage matrix's capacity to retain water within its structure. The results align with Riernersman *et al*. (2016) where the addition of chia seeds in meat products improved their water retention by up to 92 %, thus improving the WHC. According to Gore *et al*. (2022), the fibre source added to sausages tends to absorb the moisture from the protein fraction, causing aggregation of myofibrillar protein, leading to higher WHC. The reduced moisture content of the CFS samples created a medium with a higher ionic strength, favouring protein extractability and improving the binding quality of sausages (Carballo *et al*., 1995). Nevertheless, the key to developing good-quality fish sausages depends on the initial raw material quality. Even though the fish muscle contains 60 – 75 % of myofibrillar proteins, temperature changes can affect the natural configuration of proteins, leading to denaturation and decreased gel-forming and WHC (Nair, 2005). Therefore, it is essential to maintain the fish in the cold chain right from the catch to obtain stronger emulsions from it.

3.3.3 Reduction in cooking loss

It was observed that the cooking loss of the samples also recorded a proportionate decrease with the increased level of chia seed inclusion (Fig. 2 c). The CS samples exhibited a cooking loss of 10.81 % compared to 7.46 %, 5.85 % and 4.32 % for the CFS 2 %, 4% and 6 % samples, respectively. The results can be explained by the viscous structure of the polysaccharides in chia seeds, which reduces cooking loss by preventing the loss of water as described by Bushway *et al*. (1981). The improved WHC of CFS samples further contributed to the better cooking yield of the product. The result aligns with Alvarez *et al*. (2017), who reported better cooking characteristics for the restructured meat

of common carp substituted with chia flour. A perfect negative correlation was observed between WHC and the cooking loss of the sausage samples (r = -0.98; p=0.001). The hydrogen bond formation between the fibre and water molecules in the matrix results in increased water retention and a reduction in cooking loss of products (Ang, 1993).

3.3.4 Total Expressible Fluid (TEF)

The Total Expressible Fluid (TEF %) values are shown in Fig. 2. d. Statistically significant differences were noticed between the TEF % values of all the formulations ($p<0.05$). The CS sausages showed higher TEF % (29.66 %) compared to the chia-substituted sausages. A higher TEF % value indicates more water and fat release from the product during centrifugation, leading to unstable emulsions. However, with the addition of chia seed, the emulsion stability significantly improved compared to the CS samples ($p<0.05$). The lowest TEF percentage of 17.51 was achieved by the CFS 6% samples. A high negative correlation was observed between the TEF % and the increase in chia seed inclusion (r = -0.97). A higher fat percentage of the CFS 6% samples resulted in a denser and more concentrated continuous phase of the emulsion, leading to a structure with high WHC (Carballo *et al*., 1995). Furthermore, Coorey *et al*. (2014) found that chia seeds outperform commercially available gelatin and guar gums in terms of their ability to emulsify. Moreover, the total emulsion stability of the finished product is also influenced by the protein components found in chia seeds, such as albumin, glutelin, and globulin (Arifin *et al*., 2021)

3.3.5 Folding test

The folding test scores, with a grade above 3 for CS and CFS samples indicated the structural integrity of the sausages prepared from *G. suppositus* (Table 2). A significant difference was observed between the folding test scores of CS and CFS 6 % samples ($p<0.05$). The incorporation of chia seeds improved the foldability of the sausages up to grade 4 for CFS 4% and grade 5 for CFS 6% samples. The higher grade for CFS samples can be correlated well with the cohesiveness and springiness values of the sausage samples. The inclusion of chia seed flour in sausages produced higher folding test grades, contrary to the substitution with oat fibre (Gore *et al*., 2022) and mocaf flour into sausages (Elang *et al*., 2018) where the product showed lower folding test grades compared to the control. This can be attributed to the high water-holding and oil-holding capacity of the gum present in chia seeds, functioning as emulsifiers and stabilizers which contributed to improving the overall texture profile and foldability of the resultant sausages (Segura-Campos *et al*., 2014).

3.4 Physical properties of sausages

3.4.1 Colour parameters

The L*, a* and b* values for the control and fortified sausages are shown in Table 2. The L* recorded the highest value for CS samples compared to chia-incorporated sausages. With the increase in chia seed inclusion, the sausages developed a dark colour, contributing to the decrease in lightness values. The lightness values decreased from 57.83 for CS samples to 40.49 for the CFS 6 % samples. A similar reduction in L* values was observed in bread crumbs added with chia seeds (Romankiewicz *et al*., 2017). The a* exhibited a value of 1.71 for the CS samples. However, the inclusion levels of chia seed brought significant changes in the redness value for the fortified sausages compared to the CS samples. The redness values were the highest for CFS 6% samples (3.25). Similarly, the addition of chia seeds caused an increase in a* values in chicken nuggets

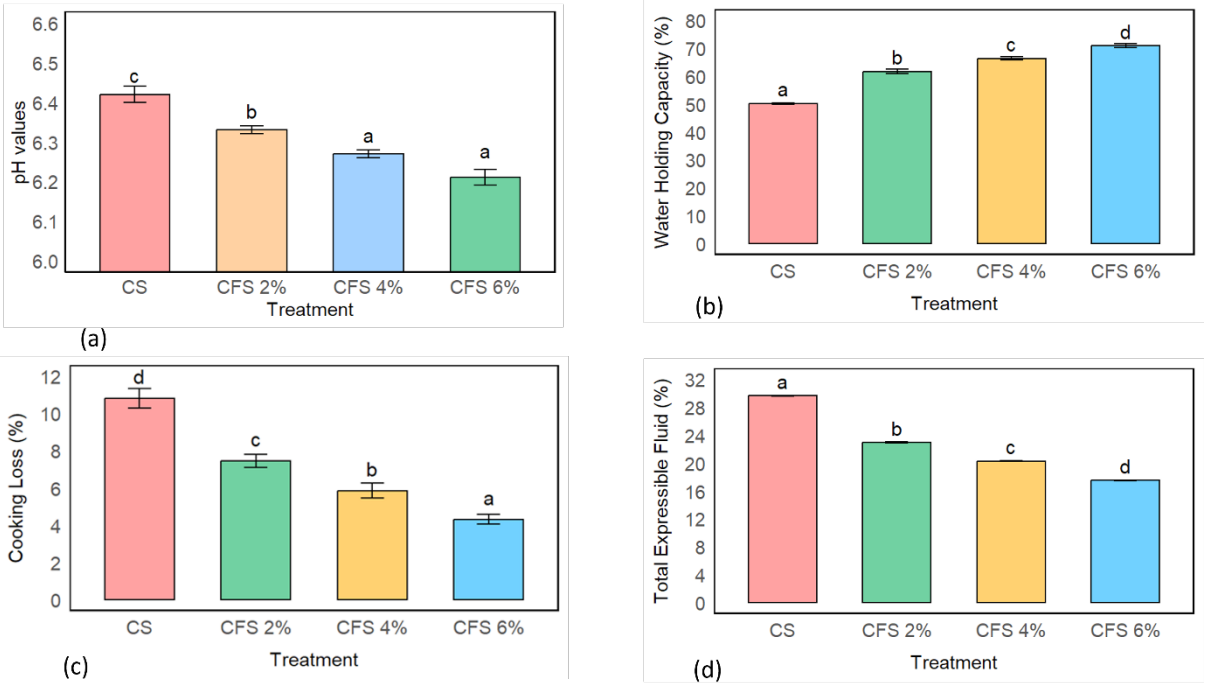


Fig. 2. (a) pH, (b) Water holding capacity, (c) Cooking loss, (d) Total expressible fluid content of CS and CFS sausages expressed as mean ± standard deviation (n=3); $p<0.05$

(Barros *et al*., 2018) and an increase in a* and b* values for the restructured common carp meat fortified with 8 % chia seeds (Alvarez *et al*., 2017). However, in the current study, with the increase in the a* values, there was a significant decrease in the b* values ($p<0.05$). While CFS 2% and CFS 4% samples did not show any significant difference in b* values, statistical differences were significant between the b* values of CS samples to those of fortified samples.

3.4.2 Textural properties

The Texture Profile Analysis (TPA) of sausage samples are given in Table 2. A significant difference was observed between the hardness values of CS and CFS sausages. This might be due to the increased surface area for interaction between the hydrophilic protein molecules and the soluble fibre, leading to a softer texture in the CFS sausages compared to the control, similar to the observation by Arifin *et al*. (2021). Among the CFS samples, CFS 6% samples exhibited the lowest hardness values. Cohesiveness values increased with the increase in chia seed addition, with a significant difference observed between the CS and CFS 6% samples. Regarding the springiness values, the highest values were recorded for CFS 6% samples, significantly different from CS, CFS 2% and CFS 4% samples. Notably, the springiness index for CFS 6% was close to 1 which indicated that the product almost retained back to shape without deformation. However, the gumminess values were significantly reduced with the addition of chia seed in sausage samples. Similar reductions in gumminess of wheat bread fortified with chia seeds were observed by Romankiewicz *et al*. (2016). The chewiness values of CFS samples decreased with chia seed addition indicating that the ease of chewiness was the highest for CFS 6% samples. Similarly, Elavarasan *et al*. (2024) observed a decrease in chewiness for frankfurters fortified with chia seeds. Similarly, fracture force increased with chia seed addition whereas a reduced stiffness was observed with increasing the amount of chia seeds.

3.5 Sensory analysis of the developed sausages

The sensory scores for the sausage formulations are represented in Fig. 3. The CS samples prepared from the low commercial value fish, *G. suppositus,* recorded a mean overall acceptability score of 7.40, which confirmed the acceptance and suitability of *G. suppositus* in sausage production. There was a significant difference between the sensory scores of the colour attribute in all four formulations ($p<0.05$). The colour of CS samples scored the highest mean value (8.23), whereas, CFS 6% samples recorded the lowest mean average score of 3.13. Similar to the observations, Nakov *et al*. (2024) claimed that adding chia seeds to yoghurt had a detrimental impact on its appearance. Regarding the odour, comparable scores were recorded for each formulation ranging from 6.7 to 8.1, with CFS 6% samples recording the highest value (8.1). The CFS 2% and CFS 4 % samples did not show any significant difference between the odour values whereas a significant difference in the mean odour scores was observed between the CS and CFS 6 % samples ($p<0.05$). The CFS 6% samples displayed a flour-cake texture due to the reduction in fish meat concentration with the lowest mean value of 5.49 in comparison to the highest mean score of 8.4 for the CFS 2% sausages. Regarding the flavour attribute, there was no significant difference between any of the samples ($p>0.05$). The CS samples recorded a mean flavour score of 6.7. The CFS 2% samples demonstrated an equal blend of fishy taste as well as nutty taste of chia with a mean score of 7.9. The CFS 4% samples recorded the highest mean average score of 8.4 whereas the CFS 6% samples recorded the lowest value (5.7) for the flavour attribute. Even though a reduced score for colour attributes was observed for the chia-fortified sausages, overall acceptability was higher for the CFS 4 % samples among all the formulations with a mean score of 4.6, 7.96, 7.9, 8.4 and 9.1 respectively for colour, odour, texture, flavour and overall acceptability.

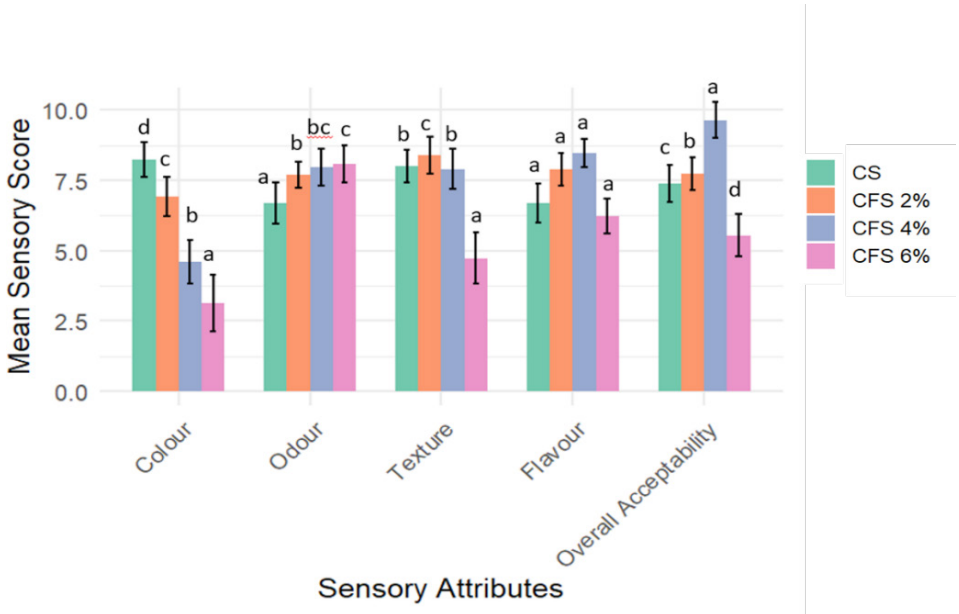


Fig. 3. Sensory scores of the CS (Control sausage) and CFS (Chia-fortified sausage) formulations expressed as mean ± standard deviation (n=3); $p<0.05$; Means with different letters in a row are significantly different.

Table 3. Fatty acid composition of *G. suppositus* sausages fortified with chia seeds (g/100g)

Fatty acids	Structure	CS	CFS 4 %
Pentadecenoic acid	C15:1	N.D	0.10 ± 0.35
Palmitoleic acid	C16:1	N.D	0.17 ± 0.07
Heptadecenoic acid	C17:1	N.D	0.26 ± 0.02
Elaidic acid	C18:1 t	0.14 ± 0.05	0.57 ± 0.02
Nervonic acid	C24:1 n-9	0.32 ± 0.02	0.27 ± 0.37
	Σ MUFA	0.46 ± 0.09	1.37 ± 0.15
Linoleic acid	C18:2 n-6	0.27 ± 0.34	0.67 ± 0.17
Linolenic acid	C18:3	0.37 ± 0.43	0.54 ± 0.05
Eicosadienoic acid	C20:2	N.D	0.61 ± 1.34
Docosahexanoic acid	C22:6 n-3	0.25 ± 0.24	0.22 ± 1.73
	Σ PUFA	0.89 ± 0.05	2.04 ± 0.17
Behenic acid	C22:0	0.46 ± 0.07	0.50 ± 0.05
Tricosanoic acid	C23:0	0.46 ± 0.04	0.40 ± 0.06
Lignoceric acid	C24:0	0.41 ± 0.11	0.35 ± 1.32
Heneicosanoic acid	C20:1	0.43 ± 0.03	N.D
	Σ SFA	1.76 ± 0.02	1.25 ± 0.06

Data expressed as mean ± standard deviation (n = 3); Σ MUFA = Sum of monounsaturated fatty acids; Σ PUFA = Sum of polyunsaturated fatty acids; Σ SFA = Sum of saturated fatty acids; N.D= Not detected

3.6 Fatty acid profile of the sausages

The fatty acid profile of CS and CFS 4% samples are shown in Table 3. The results indicated that the incorporation of chia seed significantly improved the fatty acid profile of sausages compared to the CS samples. The high susceptibility of polyunsaturated fatty acids (PUFA) to oxidation during the processing and cooking of fishery products, results in quality degradation leading to off-odour and flavour, discolouration, slimy texture, nutrient loss and production of toxic compounds (Mohammad *et al*., 2021). To prevent these quality losses, plant-derived natural extracts are used as a safer alternative to synthetic antioxidants in food preservation (López-Pedrouso *et al*., 2022). In the study, the monounsaturated fatty acids (Σ MUFA) and polyunsaturated fatty acids (Σ PUFA) of the CFS samples were higher than the CS samples while the total saturated fatty acids (Σ SFA) showed a reduced amount with chia seed incorporation. Moreover, among the PUFA, the docosahexaenoic acid (DHA) (C22:6), linolenic acid (C18:3 n3), and linoleic acid (C18:2 n6) were significantly higher for CFS 4% samples compared to CS samples ($p<0.05$) highlighting the role of chia seeds in reducing the oxidation of sausage samples. The MUFA including C15:1, C16:1 and C17:1 was below the detection limit for CS samples compared to CFS 4% samples. The decreased level of SFA and the presence of DHA and omega-3 and omega-6 fatty acids in CFS 4% samples indicate an improvement in fatty acid profile with chia seed inclusion. The result aligns with Romankiewicz *et al*. (2017) who reported a decrease in saturated fat and an increase in α-linolenic acid compared to control for wheat bread fortified with chia seeds. The observed absence of certain fatty acids in the CS and CFS sausages can be attributed to thermal degradation, or process-related losses which warrant further investigation.

The study proves that *G.suppositus* is an alternative protein source for the reformulation of sausages. The proximate composition of the fish revealed comparable scores with commercially important species. The developed sausages recorded good nutritional value, and further fortification with chia seed enhanced their nutritional synergy, functional attributes and sensory scores. Except for the colour values, the CFS samples exhibited better sensory acceptance than CS sausages. Although the respondents found all the samples to be sensorily acceptable, the CFS 4% samples recorded the highest overall acceptability score. The PUFA and MUFA content of CFS 4% sausages improved with chia seed addition. For the development of reasonably priced and nutrient-dense final products, *G. suppositus* can be sourced as an affordable raw material. Further fortification of the product using chia seeds can enhance its nutritional quality to provide better health benefits.

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