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Millet flour as a potential ingredient in fish sausage for health and sustainability

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Flour from nine varieties of millets-finger millet, foxtail millet, little millet, kodo millet, pearl millet, proso millet, barnyard millet, browntop millet and sorghum was compared with corn flour in the formulation of fresh tilapia sausages, at 10% inclusion level. The parameters compared were proximate composition, colour, texture attributes, and sensory acceptability. Millet flour-added sausages showed a significant difference ($p < 0.05$) for most of the varieties in the biochemical constituents compared to the control. Millet flour inclusion did not affect the textural characteristics of sausages and resulted in comparable viscoelastic properties as revealed by the folding test. The colour of raw millet flour did not have any correlation with the final sausage colour. In sensory evaluation, the millet-included sausages presented higher overall acceptability scores than the one prepared using corn flour. Millet flour in fish sausage formulation was concluded to be an ideal healthy substitute to conventionally used flours in sausage and contribute to SDG-2.

Sustainability spotlight

Developing novel food ingredients and combinations which are both resource efficient and nutritionally wholesome is one of the most crucial steps towards sustainable food processing. Fish is a relatively sustainable and healthier source of animal protein which contains essential amino acids and healthy fats. Millets, being rich in dietary fiber and micronutrients, and also a sustainable crop, are currently being promoted by the UN as part of the 2030 Agenda for Sustainable Development and Sustainable Development Goals (SDGs). The combination of fish and millets can result in a healthy product with combined health benefits. The present study has attempted to introduce this rarely explored combination in the form of millet-based tilapia sausages and evaluate its nutritional and physical characteristics along with its sensory qualities. The study covers 9 varieties of millets which are commonly grown and consumed across the world and compares it with corn flour which is a conventionally used flour in sausages. The fusion of millets and fish can address multiple fronts: promoting the consumption of millets, abundant in countries like India, while also popularizing a more sustainable, healthier alternative to conventional meat-based sausages. It can also contribute to the SDG of food security in millet growing countries and can lead to socioeconomic growth by improving the livelihood of stakeholders – both in the fish and millet processing industries.

1. Introduction

Growing awareness and concern surrounding the impact of food production and consumption on the environment and health has made global food trends evolve towards sustainability and personal wellness. The development of innovative products with sustainable and nutritious ingredients is essential to meet this rising demand. As a critical source of global protein supply which accounts for 17% of total animal protein consumed, fish and other aquatic foods together termed as 'Blue foods' present an opportunity for improved nutrition with lower environmental impact.[1–3] Developing diversified products from fish mince will help and encourage consumers to include more fish in their diet by replacing conventional meats such as beef, pork and poultry which have a higher environmental footprint. Although fish is a great source of high-quality proteins with well-balanced essential amino acids, therapeutic Polyunsaturated Fatty Acids (PUFAs) like EPA and DHA, and several minerals, it is not a good source of carbohydrates and functional nutrients like dietary fibers. It would be beneficial to combine fish meat with other ingredients which could provide lacking nutrients to develop a nutritionally well-balanced product with acceptable texture and sensory quality.

Sausages are a popular food choice in today's convenience-driven lifestyle. The shift towards healthy eating has made consumers seek healthier alternatives to meat-based sausages which are commonly consumed. The preparation of fish sausage involves blending fish mince with other ingredients to obtain a homogeneous paste, which is then stuffed into casings and subjected to heat processing. They can be a healthy replacement for conventional meat-based counterparts, as fish

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is a healthier source of protein and contains lower calories.[4] Several studies have suggested that sausages have a versatile composition. This allows for the incorporation of functional ingredients which may improve its keeping quality and nutritional value, and provide health benefits.[5–11] Starch and less commonly, whole flours are used as fillers in sausages to enhance the textural characteristics as fish muscle protein alone cannot hold together such a cohesive processed product.[12] These are mostly low cost flours sourced from plants such as starch from corn, cassava and potato, and refined wheat flour There have been attempts to replace such conventional starches/flours in sausages with more functional and healthy ones derived from various alternative sources like pulses, legumes and pseudo cereals.[13–15]

Millets have recently garnered a lot of attention as the year 2023 was declared as 'International Year of Millets' by the United Nations General Assembly.[16] They are recognized as 'nutri-cereals' which can flourish in extreme conditions with minimal resources, making them a highly sustainable crop that can effectively address food and nutritional security goals. According to the data collected by APEDA for the year 2020, India is the world's largest producer of millets with the pearl and sorghum varieties together contributing to around 19% of the global production.[17] The different varieties of millets, often categorized based on size as major and minor millets, are abundant in carbohydrates, protein, dietary fiber, polyphenols and various micronutrients. The nutritional profile of millets differs significantly depending on processing, variety and cultivar.[18] Most millets have been found to be hypoglycemic with the glycemic index (GI) less than 55, and are naturally gluten-free as well, making them a good substitute to conventional cereals such as rice and wheat for people with health conditions like diabetes, celiac disease, etc.[18,19] There are many studies reporting the usage of millet flour as ingredients in bakery and extruded products and as extenders in different meat products like sausages, meatballs, patties, and nuggets.[20–28]

Studies have reported the use of fish flour and millet in cookie formulations. In spite of the recognition earned by millet and fish as sustainable food commodities and their contribution to sustainable development goals particularly for SDG2, from the literature available, it is clear that there exist gaps in knowledge and technology in using the millet in fish products specifically in fish sausage. Frankfurter-type sausages from red tilapia filleting waste have been formulated with varying concentrations of quinoa flour from which 10% was found to be the optimum concentration in terms of physicochemical profile, textural properties and oxidative stability on storage.[15] But there is no literature available to date investigating the effect of adding millet flour in fish mince-based products. The objective of the present study is to explore the scope of such a combination in tilapia sausage formulation, by incorporating nine different varieties of millets at a 10% inclusion level and comparing them against a control prepared with corn flour. Such an inclusion results in providing added nutritional and health benefits to the consumer. Hence, this intervention in the fish sausage industry would ultimately contribute to overcoming the different forms of malnutrition which is one of the major obstacles to achieving SDG2.

2. Materials and methods

2.1 Materials

A fresh batch of tilapia (*Oreochromis niloticus*) was procured from a fish retailer in Kochi, Kerala province, India and transported under iced conditions to the pilot scale fish processing plant of ICAR-Central Institute of Fisheries Technology (CIFT), Kochi. On arrival, fish was washed briefly in chilled potable water, followed by descaling, evisceration and filleting. The temperature during the process was maintained below 5 °C using flake ice. The varieties of millets employed in the study were the following – finger millet (*Eleusine coracana*), foxtail millet (*Setaria italica*), little millet (*Panicum sumatrense*), kodo millet (*Paspalum scrobiculatum*), pearl millet (*Pennisetum glaucum*), proso millet (*Panicum miliaceum*), barnyard millet (*Echinochloa esculenta*), browntop millet (*Brachiaria ramosa*), which are hereafter addressed as FM, FTM, LM, KM, PM, PRM, BM, BTM respectively and sorghum (*Sorghum bicolor*). The millet grains and all other ingredients were procured from different supermarkets in Kochi. The grains were powdered (<500 μm) with the help of a mixer grinder (Philips, Mixer Grinder HL 7610/04, India). The reagents and chemicals used in the analyses were of guaranteed reagent grade or analytical grade.

2.2. Tilapia sausage preparation

The process flow chart of tilapia sausage preparation is presented in Fig. 1. Tilapia fillets were fed into a meat-bone separator (Paoli, Model 22, Rockford Illinois, USA) to obtain mince and this was used for sausage preparation according to the method described by Raju *et al.* (2003).[10] The recipe followed is presented in Table 1. For comparison, a control batch of sausages was prepared with 10% corn flour. The fresh mince along with other ingredients were thoroughly blended in a prechilled bowl chopper (MTK 661 Garant, D-72175 Dornhan, Germany), keeping the total time of comminution restricted to 12 min. The prepared paste was immediately stuffed into a cellulose casing (flat width – 25 mm), with the help of a hydraulic sausage stuffer (IS V25 IDRA VER., SIRMAN, Italy). Pieces of cotton twine were used to secure casing ends. The stuffed paste along with the casing was heat processed (100 °C) for 45 min in a steamer (gas rice cooking machine, model-GM-R10) and subsequently subjected to a thermal shock by immediately chilling in an ice bath for 12 minutes and re-steaming for one minute. Then the sausages were air cooled to ambient temperature, packed and stored in a refrigerator (±4 °C) overnight before performing the analyses.

2.3. Analyses of millet flours and tilapia sausage

2.3.1 Proximate composition of millet flours and tilapia sausage. The proximate composition of millet flour as well as the sausages formulated with each millet variety was analysed according to the methods elaborated in AOAC.[29] In brief, for moisture estimation, the sample was subjected to oven drying at

Fig. 1 Process flow of tilapia sausage preparation.

Table 1 Recipe followed for tilapia sausage preparation[a]

Ingredient		Weight per 100 g
Tilapia mince		70
Salt		2
Sugar		1.5
Sodium tripolyphosphate (STPP)		0.2
Millet flour[a]		10
Water		10
Refined vegetable oil		5
Spices	Chilli	0.4
	Coriander	0.3
	Garam masala	0.2
	Pepper	0.2
	Ginger garlic paste	0.1

[a] Nine varieties of millet flours were used in individual sausage batches and hereafter referred as FM – finger millet, FTM – foxtail millet, LM – little millet, KM – kodo millet, PM – pearl millet, PRM – proso millet, BM – barnyard millet, and BTM – browntop millet.

101 ± 1 °C (Rotek Thermostat, Mumbai) which was according to AOAC 934.01. The Kjeldahl method was used for total nitrogen estimation. A conversion factor of 6.25 was applied to determine total protein content (AOAC 954.01). Total fat content was estimated through Soxhlet extraction with petroleum ether as solvent (AOAC 920.39). Total ash content was determined by incinerating the sample in a muffle furnace maintained at 550 °C (AOAC 942.05). The carbohydrate content present in the sample was estimated through the method of difference (100 – sum of the mass of other constituents including moisture, protein, fat, and ash).

2.3.2 Colour analysis of millet flours and tilapia sausage. The colour of millet flours and tilapia sausages was measured by HunterLab (Color Flex EZ, USA) using Star Lab software. The sample cups were evenly filled with the flours for measurement. The sausages were brought to ambient temperature and blended into paste and a sample cup was filled with this paste, evenly covering the bottom area. The instrument was first standardized using two ceramic tiles – a black tile followed by a white tile before measurements were taken. The results were recorded as $L*$, $a*$ and $b*$ where $L*$ denoted lightness; $a*$ denoted degree of greenness/redness where $-a* =$ greenness and $+a* =$ redness; and $b*$ for blueness/yellowness where $-b* =$ blueness and $+b* =$ yellowness. A total of 3 replicates were measured for each sample. Additionally, for a better assessment of colour, the two mentioned values were combined to obtain the chroma values according to the equation, chroma $= \sqrt{(a*)^2 + (b*)^2}$.

2.3.3 Texture analysis of tilapia sausages. The refrigerated sausage samples were brought to ambient temperature and the casing was peeled off. The textural properties of the samples were determined using a texture analyzer (Lloyd Instruments Ltd, West Sussex, UK) through Texture Profile Analysis (TPA) and gel strength determination. Cylindrical samples of 3 cm height and 2.3–2.5 cm diameter were tested using a 500 N load cell. A total of 15 replicates from each batch of sausages were taken for the tests.

2.3.3.1 Texture profile analysis (TPA). The sample was loaded onto the sample stage where it underwent double compression to 65% of its initial height by a cylindrical probe of 4.5 cm diameter. The testing conditions followed for all samples were set uniformly as 1 mm s^{-1} crosshead speed and 0.05 kgf trigger force. The hardness (N), cohesiveness, springiness index, chewiness (Nmm), adhesiveness (Nmm) and stiffness (Nmm^{-1}) of the sausages were estimated from the force-deformation graphs obtained as described by Bourne (1978).[30]

2.3.3.2 Gel forming capacity. The gel strength of the samples was determined using a single hardness setup by inserting a ball probe into the sample at a speed of 1 mm s^{-1} till a depression limit of 15 mm from the trigger. The trigger force was 0.005 kgf. From the resulting graph, the deformation (mm) and peak load (gf) values were noted.

2.3.3.3 Folding test. A slice with 3 mm thickness taken from the middle portion of the sausage was folded into quadrants. The evaluation was carried out in accordance with a 5-point grade system given below. 5 – no crack occurs even if folded in four; 4 – no crack occurs if folded in two, but crack(s) occur(s) if folded in four; 3 – no crack occurs if folded in two, but splits if folded in four; 2 – cracks if folded in two; 1 – splits into two if folded in two.[31]

2.3.4 Sensory evaluation. The samples were warmed in a steamer and presented in white-coloured plates to a panel of 10 non-trained members consisting of staff and students of CIFT, who have exposure to tasting fish-based products. Mineral water was distributed for rinsing the palate between samples. The sensory evaluation took place inside a specially designed sensory booth with adequate lighting. The sausages were rated for sensory parameters such as appearance, colour, odor, taste, flavour, texture and overall acceptability. The scores were based on a 9-point hedonic scale as described below.

09 – like extremely; 08 – like very much; 07 – like moderately; 06 – like slightly; 05 – neither like nor dislike; 04 – dislike slightly; 03 – dislike moderately; 02 – dislike very much; 01 – dislike extremely.

2.3.5 Statistical analysis. The collected data were subjected to descriptive statistical analysis using IBM SPSS Statistics (Version 25) software and the results are presented as mean values with standard deviation. Proximate analysis of flour and sausages was carried out in triplicate. Colour analysis and sensory evaluation were also carried out in triplicate. The sample size (n) for texture profile analysis and gel strength was 15 (5 sausage × 3 pieces). One way analysis of variance was conducted to find out the presence of significance with the alpha value of 5% (confidence level 95%). Post ANOVA, the significant differences between the mean values were determined using Duncan's multiple range comparison test and grouped accordingly.

3. Results and discussion

3.1. Proximate composition of millet flours and tilapia sausage

The proximate composition of each millet flour and tilapia sausage prepared with them is presented in Tables 2 and 3, respectively. Moisture content of the flours varied from 0.69 (little millet) to 10.35% (finger millet). Sruthi and Rao observed from previously reported values that millet flours had an average moisture content of 6–13%, which increased significantly on prolonged storage.[32] In the present study, a wide variation in the moisture content of flours was observed. It should be mentioned that since the millets used in this study were procured from commercial sellers, uniformity in processes like drying cannot be ensured. However, the moisture content of the flours could affect textural properties when used in product development through interactions with water and other macromolecules like proteins.

The moisture content of sausages ranged from 65.94% in PM sausage to 69.39% in sorghum sausage. This is similar to previously reported values for fish sausages – 64.55% (ref. 15) and 68.64% (ref. 10) but lower than what Cardoso et al.[33] reported for cod frankfurters (72.20%). The total moisture content in the sausage is determined by the combination of added water (10 g/100 g), the moisture content of raw fish (81.18% for tilapia mince used in the study), and the moisture content of the respective millet flour used. It was noted that except for sorghum and PRM sausages, all millet sausages had significantly ($p < 0.05$) lower moisture content than the control (68.19%).

The crude protein content of the millet flours was in the range of 8.41 to 16.18%. It was noted that the values differed significantly ($p < 0.05$) between each millet, with the exception of LM and KM. The highest value was recorded for BTM (16.18%) which was lower than what Santhi Sirisha et al. reported (17.31%).[34] Among millet flours, FM contained the least amount of crude protein (8.41%) which is very close to the value of 8.58% reported by Ravindran.[35] Bora et al. reported protein content in a range of 6.20–14.10% for the millets with KM and FM having the lowest values.[19] All millet flours studied had significantly higher crude protein content compared to the control which only had 1.91%. Based on the protein content, the millets studied can be arranged as BTM > FTM > PRM > BM

Table 2 Proximate composition of millet flours[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Corn
Moisture (%)	10.35 ± 0.45[a]	4.12 ± 0.39[d]	0.69 ± 0.32[f]	1.72 ± 0.09[e]	10.23 ± 0.22[a]	8.55 ± 0.35[b]	8.84 ± 0.21[b]	7.71 ± 0.80[c]	7.60 ± 0.22[c]	9.94 ± 0.58[a]
Protein (%)	8.41 ± 0.28[h]	15.06 ± 0.59[b]	11.73 ± 0.04[e]	11.31 ± 0.58[e]	10.06 ± 0.01[f]	13.60 ± 0.06[c]	12.54 ± 0.53[d]	16.18 ± 0.43[a]	9.03 ± 0.11[g]	1.91 ± 0.22[i]
Fat (%)	1.57 ± 0.02[f]	4.00 ± 0.10[c]	3.85 ± 0.14[cd]	3.04 ± 0.05[e]	4.58 ± 0.28[b]	2.93 ± 0.15[e]	3.37 ± 0.27[de]	5.27 ± 0.18[a]	3.69 ± 0.92[cd]	0.41 ± 0.04[g]
Ash (%)	1.26 ± 0.25[d]	1.28 ± 0.06[cd]	1.48 ± 0.07[bc]	0.86 ± 0.17[e]	1.52 ± 0.13[b]	1.42 ± 0.02[bcd]	1.28 ± 0.05[cd]	2.58 ± 0.04[b]	1.59 ± 0.04[b]	0.28 ± 0.07[f]
Carbohydrates (%)	78.41 ± 0.71[c]	75.65 ± 0.38[d]	82.23 ± 0.21[b]	83.04 ± 0.69[b]	73.59 ± 0.31[e]	73.48 ± 0.17[e]	73.94 ± 0.52[e]	68.24 ± 0.68[f]	78.07 ± 1.16[c]	87.43 ± 0.70[a]

[a] The values are presented as mean ± standard deviation. Different superscript letters in the same row indicate significant differences ($p < 0.05$) between the flours.

Table 3 Proximate composition of tilapia sausages (g/100 g) formulated with millet flours presented as the mean ± standard deviation[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Control
Moisture (%)	66.67 ± 0.21[def]	66.87 ± 1.06[de]	65.97 ± 0.31[f]	67.04 ± 0.35[cd]	65.95 ± 0.21[f]	67.66 ± 0.14[bc]	66.22 ± 0.46[ef]	66.91 ± 0.09[de]	69.39 ± 0.10[a]	68.19 ± 0.21[b]
Protein (%)	12.31 ± 0.62[bcde]	12.03 ± 0.21[de]	13.40 ± 0.44[a]	11.80 ± 0.19[e]	12.19 ± 0.47[cde]	12.90 ± 0.09[ab]	12.48 ± 0.40[bcd]	12.74 ± 0.10[bc]	11.07 ± 0.05[f]	12.25 ± 0.06[cde]
Fat (%)	5.73 ± 0.07[bcde]	5.99 ± 0.23[bcd]	6.41 ± 0.23[bc]	5.50 ± 0.30[cde]	7.62 ± 1.44[a]	5.35 ± 0.23[de]	6.61 ± 0.14[b]	6.36 ± 0.13[bc]	6.14 ± 0.10[bcd]	4.93 ± 0.13[e]
Ash (%)	2.93 ± 0.02[a]	2.79 ± 0.15[b]	3.01 ± 0.08[a]	2.18 ± 0.05[e]	2.63 ± 0.05[c]	2.37 ± 0.03[d]	2.41 ± 0.09[d]	2.47 ± 0.02[d]	2.10 ± 0.03[e]	2.47 ± 0.02[d]
Carbohydrates (%)	12.33 ± 0.77[b]	12.29 ± 0.73[b]	11.19 ± 0.45[d]	13.46 ± 0.12[a]	11.59 ± 0.86[bcd]	11.70 ± 0.25[bcd]	12.25 ± 0.34[b]	11.49 ± 0.18[bcd]	11.27 ± 0.02[cd]	12.14 ± 0.41[bc]

[a] Different superscript letters in the same row indicate significant differences ($p < 0.05$).

> LM > KM > PM > sorghum > FM. The major protein fraction found in millets consists of prolamines[36,37] and they are found to have low lysine contents but higher methionine content than most cereals like rice.[38,39] FM is reported to have a higher level of methionine (210 mg g^{-1} N^{-1}), compared to all other millets.[39] The measure of protein quality of a substance is indicated by its chemical score, and finger millet was reported to have 52, while PM had 63 and sorghum, 37.[40]

Among the formulated sausages, PRM (12.90%) and LM (13.40%) sausages contained significantly higher amounts of protein and sorghum had the lowest (11.08%). Apart from these, all other millet sausages exhibited similar protein content to the control, having no significant differences ($p > 0.05$) among them. Since sausages are basically emulsion-based products the functionality of the proteins involved plays an important role in the network formation during gelation. The myofibrillar proteins present in fish are mainly responsible for the product's textural characteristics. But, when millet flour is incorporated into the product, their interaction with millet proteins could result in a synergistic effect based on compatibility among these proteins. The functional properties of each millet flour need to be evaluated to predict their behavior in an emulsion. The interactions between proteins and lipids have a major role to play in the entrapment of fat in sausage batters.[41] Whole grain flour of PRM was found to have appreciable oil absorption capacity, hence holding significant potential as an ingredient in meat emulsion products.[42] Significant differences were not observed in crude protein content between millet and control sausages (except LM, PRM and sorghum).

BTM had the highest fat content of 5.27% which is lower than the 6.27% reported by Santhi Sirisha et al.[34] PM is often reported to contain higher fat content (5–7%) among other millets and also among most cereals.[18,19,36] However, these reported studies did not include BTM. The lowest fat content among millets was observed in FM (1.57%). Shobana et al.[39] also observed lower lipid content in finger millet (1.3%) in comparison to the other millets studied (FTM, LM, KM, BM and PM). Corn flour had very low fat content (0.41%). Lipid content in the millet sausages varied between 5.35% in PRM and 7.62% in PM while in the control it was only 4.93%. Hence, it is clear that inclusion of millet flour in sausage significantly ($p < 0.05$) raised its fat percentage.

BTM (2.58%) contained significantly ($p < 0.05$) higher ash content than other millet flours, and the observed value was close to 2.36% reported by Santhi Sirisha et al.[34] All the millet flours had significantly ($p \leq 0.05$) higher ash content than the control (0.28%). Muchekeza et al.[13] reported a very similar value (0.26%) for corn starch. This indicates that millet flour is more mineral dense than corn flour as the ash content in a sample is a representation of the total mineral content present. KM had the least amount of ash (0.87%) among all millet flours. Geervani and Eggum[38] reported (1.04%) ash for KM. Among sausages, LM contained the highest percentage of ash (3.02%) and sorghum contained the lowest (2.10%). But, although corn flour and millet flours had significantly different ash content, this did not reflect in sausages and the ash content of control sausages did not vary significantly (2.47%) from three millet

flours (PRM, BM, and BTM). Raju et al.[10] reported similar ash content (2.67%) in fish sausage.

Being cereal grains, carbohydrates form the major part of millet flour composition. The values ranged from 68.24% in BTM to 83.04% in KM, which contained significantly higher ($p < 0.05$) carbohydrates among millet flours. Corn flour contained the highest carbohydrate content (87.44%) among all the flours. Sorghum starch has been reported to have similar properties to that of maize starch making it suitable for use in formulated foods.[37,43] But it had a higher gelatinization temperature than maize starch, which could affect the cooking time.[37] Among the sausages, millet-included sausages had similar carbohydrate content to the control and only KM differed significantly ($p < 0.05$), having the highest value observed (13.46%). It should be mentioned that carbohydrate was estimated through the method of difference as in this case, includes fiber and other components like organic acids. Although the carbohydrate, protein and fat content of the flour used are important parameters, it is their unique interactions with the fish muscle that influence the gelation and the textural characteristics of fish sausage.

The proximate analysis of millet-included sausages reveals that they do not vary much from the control formulated with corn flour with certain exceptions. The compositional differences between the millet flour and corn flour are reflected in the fat, protein, ash and carbohydrate content of the tilapia sausages.

The observed significant difference in proximate compositional parameters of the sausage added with certain varieties of millet shows that preferences can be made based on the need. For example, little millet-added sausage resulted in 9% more mean protein content (13.40%) compared to the control (12.25%). Similarly, for addressing mineral deficiencies, little millet-added sausage is more preferable as indicated by the high ash content. So, for alleviating issues like malnutrition including protein deficiency, little millet-added sausage could be a better choice. On the other hand, the glycemic index (GI) and glycemic load (GL) of corn flour is 70 and 53.80, respectively. The GI value of fish is zero as it is mainly rich in protein content. Fish sausage is a low glycemic food with a GI value closer to 30 and a GL value closer to 1 mainly because of the addition of starch sources like corn flour. Hence, the sausages developed using millets could also find a better place in the therapeutic food choices of diabetic patients with still lesser GI and GL. According to the Sustainable Development Goals Report 2023, under SDG-2 (zero hunger), in the year 2022, 148 million children under five years of age had stunted growth. The major reasons are inadequate nutrition, and poor nutrition intake, utilization and adsorption. Fish sausage would be a better choice for children. Feeding children with fish has to be always under the supervision of adults due to the risk of consuming spines and pin bones. This issue is completely freed when it is given in the form of fish sausage as it is prepared only using spine-free fish meat. In this connection, development of fish sausage with millet flours and its acceptance by consumers can bring a fundamental shift in the trajectory of SDG-2.

Table 4 Color coordinates $L*$, $a*$ and $b*$ and the chroma values of the flour presented as mean ± standard deviation[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Control
$L*$	65.60 ± 0.19[h]	71.60 ± 0.03[e]	68.16 ± 0.03[g]	70.89 ± 0.02[f]	64.56 ± 0.02[i]	65.60 ± 0.19[h]	72.55 ± 0.16[d]	73.22 ± 0.02[c]	82.95 ± 0.04[b]	91.38 ± 0.03[a]
$a*$	2.13 ± 0.01[b]	2.93 ± 0.02[a]	−0.33 ± 0.01[f]	2.14 ± 0.01[b]	0.02 ± 0.02[e]	2.13 ± 0.01[b]	0.94 ± 0.09[d]	1.00 ± 0.01[c]	−0.34 ± 0.00[f]	−1.71 ± 0.01[g]
$b*$	6.27 ± 0.04[h]	26.61 ± 0.04[a]	22.33 ± 0.03[b]	15.90 ± 0.02[d]	12.78 ± 0.04[f]	6.27 ± 0.04[h]	17.53 ± 0.29[c]	15.08 ± 0.01[e]	11.93 ± 0.03[g]	4.65 ± 0.03[i]
Chroma	6.62 ± 0.04[h]	26.77 ± 0.04[a]	22.33 ± 0.03[b]	16.04 ± 0.02[d]	12.78 ± 0.04[f]	6.62 ± 0.04[h]	17.56 ± 0.30[c]	15.12 ± 0.01[e]	11.93 ± 0.03[g]	4.95 ± 0.03[i]

[a] Different superscript letters in the same row indicate significant differences ($p < 0.05$).

3.2 Color evaluation

Color analysis results of each millet flour and added sausage are presented in Tables 4 and 5 respectively. Color is a crucial factor for consumer appeal which also has the ability to affect other sensory properties.[44] About 60% of the consumers were willing to purchase meat-based frankfurter-type sausages when L^* values were between 62.3 and 68.5.[45] Among the prepared sausages including control, only sorghum sausage had the L^* value in the aforementioned range, which was also significantly higher than that of the other sausages. The lowest lightness value of 50.83 was recorded for FM which reflected well in its physical appearance, being the darkest among all sausages. It is common to grade the color of meat products by its redness (a^*) value which is reported to have a high correlation ($r = 0.947$) with L^* values.[45] As in the case of L^* values, the lowest a^* value (17.10) was recorded for FM sausage. BTM and FTM sausages had significantly higher a^* compared to other sausages. The sausages prepared exhibited mean b^* (yellowness) values in the range of 3.13 (PM) to 5.29 (FM) and presented a significant difference ($p < 0.05$) among them except LM and PRM. The chroma values of the sausages ranged from 17.90 (FM) to 23.03 (FTM). Higher chroma values are associated with higher pigment concentration and increased perception of color intensity and as the values decreased, the samples became darker.[46] In cooked ground beef patties, a post-cooking time before evaluation was found to reduce a^* and b^* values while increasing brown color scores.[47] In this study, the prepared sausages were stored in a refrigerator for 18 h and steamed for 1 min before evaluation, which may have affected the measured values. The heme pigment present in myoglobin is responsible for the color of fish muscle but oxidation during heat processing significantly alters the color of the final product. Millet grains have varying quantities of pigments and other compounds imparting each variety a unique color. This has resulted in wide variations in the color of sausages prepared from different millet flours. These pigments present in millet flour were found to be sensitive to pH change.[48] The major pigments present in millet grains include carotenoids (lutein and zeaxanthin), anthocyanins, tannins and flavonoids which may contribute to the color of the sausages.[49–51]

The L^*, a^*, b^*, and chroma values of raw millet flours and sausages had no correlation. Other ingredients in the formulation such as chilli powder and black pepper and the level of oxidation of myoglobin in fish muscle during heating can also influence the color of sausages. Polyphenols and other natural antioxidants extracted from plants are increasingly becoming popular in meat and fish products owing to their positive effect on color stability and myoglobin oxidation.[52] However, there is no such specific literature on the effect of millet polyphenols in fish mince-based products and this could be the future direction of research to understand more about the color attributes of millet-added fish sausages. Meanwhile the pro-oxidative role of salts and metals present in the millets cannot be ignored. In terms of color, millet-included sausages have exhibited significantly different values from the control but its influence on

Table 5 Color coordinates L^*, a^* and b^* and the chroma values of millet flour-included tilapia sausages presented as mean ± standard deviation[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Control
L^*	50.83 ± 0.02[i]	55.23 ± 0.03[h]	59.17 ± 0.02[d]	55.62 ± 0.02[g]	56.22 ± 0.05[f]	59.19 ± 0.04[d]	60.08 ± 0.03[c]	60.40 ± 0.07[b]	63.88 ± 0.06[a]	56.64 ± 0.02[e]
a^*	5.30 ± 0.01[a]	4.55 ± 0.02[c]	4.36 ± 0.02[d]	3.22 ± 0.02[h]	3.13 ± 0.01[i]	4.37 ± 0.01[d]	3.64 ± 0.01[g]	3.53 ± 0.02[g]	4.09 ± 0.01[e]	4.79 ± 0.02[b]
b^*	17.10 ± 0.18[g]	22.58 ± 0.04[a]	21.46 ± 0.05[b]	19.05 ± 0.05[f]	20.32 ± 0.02[d]	21.45 ± 0.02[b]	20.53 ± 0.01[c]	22.55 ± 0.06[a]	20.14 ± 0.06[e]	20.53 ± 0.05[c]
Chroma	17.90 ± 0.17[h]	23.03 ± 0.03[a]	21.89 ± 0.05[c]	19.32 ± 0.05[g]	20.56 ± 0.02[f]	21.89 ± 0.02[c]	20.85 ± 0.06[e]	22.83 ± 0.06[b]	20.56 ± 0.07[f]	21.08 ± 0.05[d]

[a] Different superscript letters in the same row indicate significant differences ($p < 0.05$).

Table 6 TPA parameters of tilapia sausage formulated with millet flour are presented as mean ± standard deviation[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Control
Hardness (N)	44.27 ± 10.45[bc]	56.59 ± 6.59[a]	38.24 ± 7.43[cd]	40.45 ± 4.99[bcd]	34.64 ± 3.65[d]	43.26 ± 4.71[bc]	40.46 ± 4.44[bcd]	44.42 ± 4.45[bc]	44.64 ± 4.67[b]	35.29 ± 9.31[d]
Cohesiveness	0.17 ± 0.03[d]	0.28 ± 0.02[abc]	0.30 ± 0.01[a]	0.28 ± 0.02[abc]	0.26 ± 0.03[bc]	0.30 ± 0.01[ab]	0.25 ± 0.02[c]	0.27 ± 0.02[c]	0.27 ± 0.03[abc]	0.25 ± 0.07[c]
Chewiness (Nmm)	6.16 ± 1.94[d]	11.95 ± 1.53[a]	8.35 ± 1.63[bc]	8.44 ± 1.34[bc]	6.72 ± 1.20[cd]	8.98 ± 1.39[b]	7.33 ± 0.96[bcd]	9.07 ± 1.61[b]	8.97 ± 1.50[b]	7.42 ± 3.47[bcd]
Springiness index	0.78 ± 0.05[a]	0.73 ± 0.02[b]	0.72 ± 0.05[b]	0.72 ± 0.03[b]	0.73 ± 0.01[b]	0.69 ± 0.05[b]	0.71 ± 0.05[b]	0.73 ± 0.02[b]	0.73 ± 0.04[b]	0.78 ± 0.05[a]
Adhesiveness (Nmm)	0.82 ± 0.46[b]	0.33 ± 0.23[b]	0.94 ± 0.96[b]	0.69 ± 0.59[b]	1.64 ± 0.62[a]	0.41 ± 0.14[b]	0.68 ± 0.54[b]	0.72 ± 0.72[b]	0.54 ± 0.39[b]	0.90 ± 0.77[b]
Stiffness (Nmm⁻¹)	4.94 ± 1.66[bcd]	7.77 ± 1.40[a]	7.91 ± 3.77[a]	5.33 ± 1.00[bcd]	3.93 ± 0.94[d]	6.20 ± 1.76[abc]	4.85 ± 1.01[cd]	6.80 ± 1.43[ab]	5.49 ± 1.25[bcd]	5.07 ± 2.80[bcd]

[a] Different superscript letters in the same row indicate significant differences ($p < 0.05$).

consumer acceptance needs to be assessed through large scale acceptability studies.

3.3 Textural properties of millet-incorporating tilapia sausages

3.3.1 Texture profile analysis (TPA). The mean values of textural parameters for each group of sausages recorded during TPA are presented in Table 6.

Among the sausage samples studied, the hardness values of PM, LM, KM, and BM did not exhibit a significant difference ($p > 0.05$) from the control. According to hardness values, the millet-included sausages are in the following order – PM < LM < KM < BM < PRM < FM < BTM < sorghum < FTM. Hardness is the peak force recorded under the first compression during TPA. It is considered as an important factor in determining the consumers' willingness to purchase sausages. Dingstad et al.[45] studied the correlation between consumer acceptance and the firmness value of frankfurter-type sausage and reported that the firmness values of 40.4 N and 47.3 N as satisfactory and lower acceptable firmness, respectively. In the present study, sensory analysis revealed the best acceptance of BTM sausage which had a hardness value of 44.42 N. FTM had significantly ($p < 0.05$) higher hardness (56.59 N) compared to other sausages. The hardness of sausages is a manifestation of factors such as the nature of interaction between the protein and molecules in millet flour and the myosin network. Often the protein content of sausages is directly related to their hardness.[53,54] Increased hardness of sausages is also attributed to smaller fat globule size in the sausage gel network.[55] The protein content of tilapia used in the present study is the same (all the sausage samples were prepared using 70% meat) but the millets had different proteins in terms of intrinsic nature and quantity. There are no reports available on the effect of millet proteins on fish myosin network formation during heat-assisted gelation. The moisture content is another parameter which can influence the hardness values. The results in this study reveal no correlation (data not presented) between the major chemical constituents like moisture and protein, and moisture and hardness values. It should be mentioned that the millets are known for the presence of dietary fibers which promote and strengthen the interactions between the various components present in the fish sausage matrix. Devatkal et al.[20] reported that the addition of 10% sorghum flour increased hardness in chicken nuggets compared to those formulated with refined wheat flour (5%). The authors attributed this to the high water and fat absorption of sorghum flour which facilitates protein and starch network formation in the meat system. Dincer and Cakli[54] reported hardness values of 46.45 N and 50.65 N for trout and saithe sausages, respectively.

The cohesiveness attribute in TPA measurement is indicative of the extent of deformation of the sample by the teeth before breaking. Szczesniak[56] defines cohesiveness, as the strength of internal bonds making up the body of the product. In terms of cohesiveness, FM had the lowest value (0.18) which was significantly ($p < 0.05$) lower than the rest of the sausages including control, and LM had the highest cohesiveness value

of 0.305. Frankfurter-type red tilapia sausage with 20% quinoa flour exhibited the same cohesiveness value of 0.305. Products with high cohesiveness tend to be springier and fracture into larger fragments.

Springiness is defined as the rate at which a deformed piece of food returns to its original form.[31] FM and the control had similar springiness values which were also significantly higher ($p < 0.05$) than others. Springiness in the sausages ranged from 0.69 (PRM) to 0.78 (FM and control). Dincer and Cakli[55] reported a springiness value of 0.50 in trout sausage and 0.84 in saithe sausage. Springiness is the only parameter which is least affected by the measurement difference.[57] As the springiness values increase, the energy required for mastication also increases.[58]

It has been observed that springiness and cohesiveness values provide identical structural information about the food material, as they both serve as measures of plastic deformation.[59]

Chewiness is the energy required to masticate a solid food product to a state ready for swallowing.[56] Being the product of hardness, cohesiveness and springiness values, chewiness most often follows the trend of these values.[5] However, the results of the present study have no such correlation. This could be because of the inherent difference in the properties of the protein network formed in tilapia sausages in interaction with macro and micro molecules of the millet matrix. This needs further investigation for a better understanding of textural manifestation by millet constituents through interaction with fish myosin. FTM showed the highest chewiness (11.95 N) which is significantly higher ($p < 0.05$) than that of all other sausages while FS had the lowest value (6.16 N). Dincer and Cakli[55] reported higher chewiness values of fish sausages (18.74 N mm and 15.63 N mm for trout and saithe respectively), while Santana et al.[60] obtained a lower value (5.59 N) for surimi sausage. Lago et al.[61] and Pietrasik[62] reported lower values of chewiness in sausages containing higher fat content. A high-fat product is often soft and succulent, so consequently less chewy.[61]

Adhesiveness is defined by Szczesniak[56] as the work required to overcome the attractive forces between the food surface and the surfaces it touches during mastication, like teeth, inside of the mouth, tongue, etc. Ideally, for sausages, it is preferred to have lower values for this parameter to present a smooth and firm texture with minimum adherence to mouth parts.[61] PM exhibited a significantly higher value (1.63 Nmm) for adhesiveness compared to the rest of the sausages which did not vary significantly from each other ($p < 0.05$). FTM had the lowest adhesiveness (0.33 Nmm). The earlier reports often correlated the high adhesiveness of the material with low moisture, whereas products exhibiting a low degree of adhesiveness could be found across the entire moisture scale.[56] PM, which displayed the highest adhesiveness, also had the lowest moisture content among all the sausages. In a study conducted on Pangas mince sausage, the authors related lower adhesiveness values to lower emulsion ability which causes moisture loss on compression during TPA.[6] However, one cannot ignore the drying of the surface due to the difference in measurement

Table 7 Breaking force (g), deformation (mm) and gel strength (gmm) of tilapia sausages formulated with millet flours expressed as mean ± standard deviation[a]

Parameter	FM	FTM	LM	KM	PM	PRM	BM	BTM	Sorghum	Control
Breaking force (g)	192.98 ± 51.78[e]	229.84 ± 17.86[d]	283.02 ± 23.96[b]	262.66 ± 22.35[bc]	262.66 ± 39.87[a]	240.65 ± 31.42[cd]	236.19 ± 32.83[cd]	243.71 ± 21.41[cd]	234.54 ± 25.55[cd]	244.22 ± 30.53[cd]
Deformation (mm)	8.59 ± 0.63[d]	6.31 ± 0.81[e]	6.94 ± 0.43[e]	8.69 ± 0.93[cd]	8.99 ± 0.97[bcd]	9.49 ± 0.61[b]	8.83 ± 0.64[bcd]	9.41 ± 0.54[bc]	9.07 ± 0.65[bcd]	10.82 ± 0.87[a]
Gel strength (g mm)	1641.23 ± 350.04[ef]	1447.37 ± 189.48[f]	1968.62 ± 246.37[de]	2797.07 ± 469.18[a]	2366.10 ± 376.70[bc]	2295.37 ± 415.45[bcd]	2098.09 ± 396.74[cd]	2299.37 ± 298.25[bcd]	2129.62 ± 304.71[cd]	2638.19 ± 352.03[ab]

[a] Different superscript letters in the same row indicate significant differences ($p < 0.05$).

conditions, which can cause variation in measured adhesiveness values.

Stiffness is calculated as the maximum gradient of the force-deformation curve generated during the first compression. It denotes the highest rate of change in force with respect to deformation for the given compression strain. The stiffness values of the sausages ranged from 3.93 N mm^{-1} (PM) to 7.91 N mm^{-1} (LM). Tuna sausages containing 5% each of wheat and oats fiber presented a stiffness value of 6.72 N mm^{-1}.[7]

It is worth noting that TPA parameters are subjected to change significantly with variations in compression strain and crosshead speed used in the test. The present results were obtained on a set of test conditions selected through trials with no breakage of the sample.

TPA results of the sausages reveal that millet sausages exhibit similar textural properties to those of control on instrumental imitation of mastication, which is an indicator of textural acceptability of these sausages.

3.3.2 Gel strength. The breaking force (g), deformation (mm) and gel strength (g mm) of the sausages are presented in Table 7. Gelation is a crucial functional property of the ingredients, which affects the final product texture and acceptability, especially in sausages. The gelling quality of minced meat products is mainly evaluated by measuring their gel strength.[63] The texture analyzer measures the breaking force (similar to hardness) and deformation during a puncture test and the product of these two parameters is represented as the gel strength of the given material. The least value (192.98 g) of breaking force recorded for FM indicates weaker resistance to probe penetration which points to the weaker protein–protein network and interaction of protein with other molecules like starch. KM had a significantly ($p < 0.05$) higher breaking force of 321.72 g. The breaking force of other sausages was between 229.84 g and 262.66 g. The mean deformation value of millet sausages fell in the range of 6.30–9.49 mm. Control sausage exhibited the highest value of 10.82 mm. The millet sausages had significantly ($p < 0.05$) lower deformation values than the control but had similar breaking force values which resulted in similar gel strength. However, the deformation values imply that corn flour better preserved the gel-forming capacity of tilapia muscle proteins compared to the millet flour. Although FTM sausage exhibited the highest hardness and chewiness values among all sausages, in terms of gel strength, it had the least value (1447.37 g mm). Nagaprabha and Bhattacharya[64] investigated the gel-forming ability of foxtail millet flour alone

and found the formation of acceptable gels in deionized water at 11% concentration after subjecting to heat (95 °C for 45 min). This was attributed to the starch content (around 76%) in the millet. The addition of starch in meat emulsions results in the formation of a more compact and robust heat-induced protein network. The interactions between the fish muscle proteins and millet flour components have been rarely studied and understood with reference to the gel forming capacity of fish muscle protein. The gelation characteristics of myofibrillar proteins and the formation of a 3D protein gel network are influenced by several factors, including the nature of myosin, the source of muscle, the rate and temperature of heating, ionic strength, pH, and fat content.[65]

3.3.3 Folding test. Folding test results of the sausages are displayed in Fig. 2. The folding test is commonly carried out to judge the quality of surimi gels and can be a great tool in predicting the textural quality of gelled food products like sausages.[30] The test revealed that all the millet-incorporating sausages scored the highest grade of quality (5), similar to control sausage, which is an indicator that corn flour can be successfully substituted with millet flour without compromising on textural quality.

3.4 Sensory evaluation

The mean values of the sensory parameters scored on a 9-point hedonic scale are displayed in Fig. 3. The panellists expressed that FTM sausage was chewier than the rest and as a result, less appealing. This was supported by the higher hardness and chewiness, and lower adhesiveness values in TPA analysis. The deep red coloured appearance of FM due to the pigments present in FM might be the reason for its lowest score in appearance (8.05) among the other millet sausages. In terms of overall acceptability, flavour and texture, all the millet sausages outperformed the control, with the highest score for overall acceptability given to BTM (8.61). BTM also had the highest mean values in all the other sensory attributes. The higher scoring in flavour and texture for millet sausages in comparison to control, reveals that millet flour addition, irrespective of the type used, enhanced the organoleptic qualities of tilapia sausage.

The overall practical implications of the present study could be projected in a larger perspective as briefed here. The sustainable development goal 2 is 'to end hunger, achieve food security and improve the nutrition and promote sustainable

Fig. 2 Folding test results of the 9 millet included sausages (starting from left– FM, FTM, LM, KM, PM, PRM, BM, BTM, and sorghum), and control sausage (extreme right).
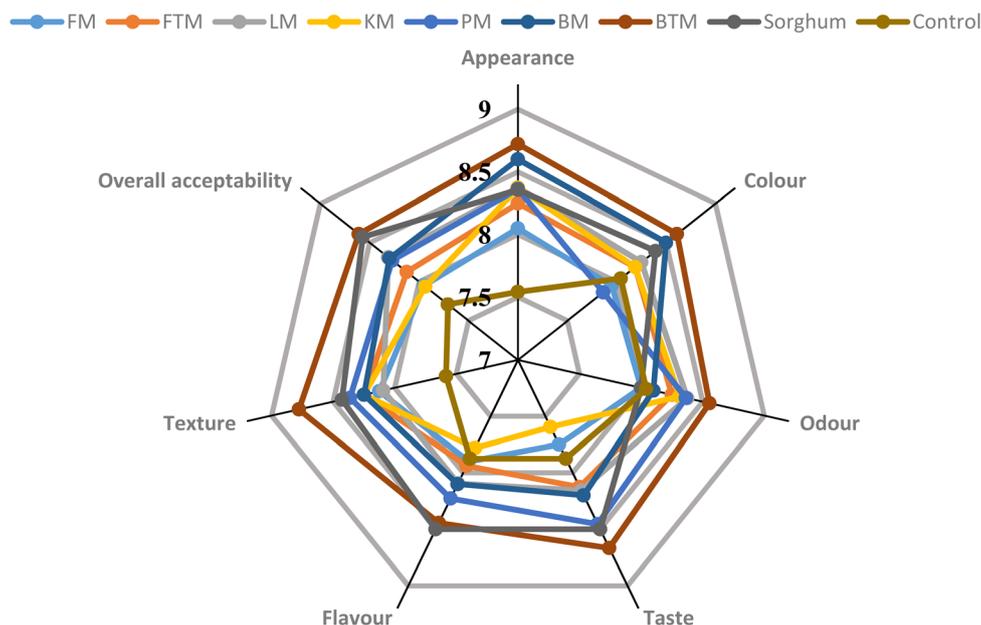
Fig. 3 Sensory attributes of the sausages evaluated on a 9-point hedonic scale.

agriculture'. In this connection, the United Nations General Assembly (UNGA) declared 2023 as the International Year of Millets which signifies the importance of millets in transforming the food system.[66] On the other hand, irrespective of the source and culture practices, aquatic food is considered to be sustainable mainly due to less carbon emission compared to land-based animals. Fish is also rich in protein, therapeutic fats and macro and micronutrients.[67] There are targets-ending hunger by providing nutritious foods, ending all forms of malnutrition by addressing the nutritional needs, and doubling the income of small-scale food producers, and fishers through value addition. Introducing healthy ingredients like millet flour in fish sausage formulation aligns well with SDG-2. Implementing such processing technology which uses sustainable ingredients, and producing and marketing at a commercial scale would largely benefit the consumers.

4. Conclusion

The proximate analysis of millet flours and sausages indicates that both millet sausages and corn flour-included sausages presented a similar nutritional profile. The presence of health-promoting phytochemicals, minerals and dietary fibre in millets makes millet added fish sausages as a healthier alternative. The study also concludes that the addition of millet flours, irrespective of variety could provide similar textural properties to that of fish sausages made with corn flour. The colour coordinates and chroma values of raw millet flour do not affect the sensory appeal of the sausages. Sensory evaluation revealed a highly positive response towards millet sausages, and in terms of overall organoleptic quality, these were superior to corn flour-based fish sausages. The nine varieties of millets used in this study proved to be equally good replacements to conventional starches in sausage preparation with added

benefits like improved nutritional quality, bioactive potential and sensory acceptability. This study forms the first report on the combination of fish mince with millet flour and the promising results obtained pave the way for future research on millet based composite fish products development.

Further studies in a similar line would be explored to understand the additional factors that may influence the quality and acceptability of millet-based sausages such as processing techniques like microwave processing, and retorting. Similarly, the storage stability under short-term (refrigeration) and long-term preservation (freezing and frozen storage of processed sausages) needs to be assessed. Many ingredient combinations like using fish oil in millet sausage formulations also need to be explored further.

Author contributions

Elavarasan K.: conceptualisation, methodology, data curation and formal analysis, validation, supervision, writing – reviewing and editing. Malini Mathew: investigation, data collection, software, visualisation, writing – original draft. George Ninan: conceptualisation, writing – review and editing. Ravishankar C.·N.: conceptualisation, writing – review and editing. Dayakar Rao: conceptualisation, writing – review and editing.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

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