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# Effect of incorporating wheat bran as a dietary fibre source in fermented carabeef sausage<sup>#</sup>

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## Abstract

This study investigated the incorporation of wheat bran into semi-dry fermented carabeef sausages, analysing their physico-chemical properties, sensory properties and microbiological quality, with an emphasis on the impact of refrigerated storage. Utilizing buffalo meat, which is prominent in India's meat industry due to its health benefits and minimal religious consumption restrictions, this research focused on enhancing the dietary fiber content of sausages a product traditionally low in fibers. Wheat bran, known for its high fiber content and health benefits, was integrated at varying levels (6%, 8% and 10%) into sausage formulations. The treatment that included 8% wheat bran emerged as the optimum choice based on cooking yield and sensory attributes, exhibiting significant improvements in moisture retention and overall acceptability compared to the control. Over 60 days of refrigerated storage, both control and most accepted treatment sausages demonstrated a reduction in pH and moisture content, with an increase in microbial counts and indices of oxidative and nitrogenous degradation. Despite these changes, the sensory parameters of treatment sausages with 8% wheat bran were maintained at acceptable levels throughout the storage period. This study concludes that the incorporation of fibre into fermented carabeef sausages can improve sensory attributes and extend the product's shelf life, suggesting a promising approach for enhancing the nutritional profile and storage stability of meat products.

Keywords: Fermented carabeef sausage, dietary fibre, wheat bran, storage study

India leads globally in buffalo meat production, with a 2019 livestock census revealing a buffalo population of 109.85 million and carabeef production reaching 1,720,030 tonnes in 2022-2023, contributing significantly to the nation's meat industry. Buffalo meat, identified for its health benefits, lacks religious consumption restrictions, making it an increasingly popular red meat choice worldwide. Sausages, a major meat product, benefit from technological advancements in shelf-stable processing, reducing energy use and aiding industry growth. Fermented sausages, made from a mix of meat, fat and spices fermented by specific bacteria, are noted for their safety, nutritional benefits and flavour, with lactic acid bacteria playing a key role (Devine and Dikeman, 2014). Lactic acid bacteria are particularly valued in bio-preservation for their safety, dominance in the natural microbiota of various foods during storage and their long-standing use as food-grade bacteria, earning them the Generally Recognized as Safe (GRAS) status. Additionally,

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bioactive peptides may be produced during meat aging and fermentation by LAB, contributing to the meat's nutritional value (Korhonen and Pihlanto, 2003). Semidry sausages, defined by a 25-30% moisture loss during processing, have a relatively higher moisture content that makes them more prone to spoilage. To counteract this, they are fermented to a lower pH to achieve a tangy flavour, often accompanied by smoking during the fermentation cycle. Despite their shelf-stable nature, semidry sausages typically require refrigeration at 4°C for preservation.

Meat lacks dietary fibers, essential nondigestible carbohydrates such as cellulose, hemicellulose, lignin, polysaccharides and oligosaccharides. These fibers which are renowned for their nutritional, functional and health benefits, offering protection against lifestyle diseases such as obesity, certain cancers, type-II diabetes, cardiovascular diseases and bowel disorders (Ajila and Raos, 2013). Incorporating dietary fibers into meat products can enhance their physicochemical properties, nutritional profile and textural qualities while also serving as cost-effective extenders/binders/fillers and fat replacers. Wheat (Triticum aestivum L.), a staple crop worldwide, is rich in proteins, dietary fibre, vitamins, minerals and phytochemicals, contributing to a balanced diet. Wheat bran, the outermost layer of the grain, is a significant byproduct of milling, renowned for its boosted total dietary fibre content, predominantly insoluble, which exceeds 50% of its weight, emphasizing its nutritional value. This study check the effect of incorporating wheat bran as a dietary fibre source in semidry fermented carabeef sausage.

## Materials and methods

Deboned buffalo meat from the carcass of adult buffaloes was procured within five to six hour post-slaughter from the Meat Technology Unit, Mannuthy. Wheat bran was directly incorporated into the sausage formulation. The condiment blend was created by combining chopped onion, peeled garlic and chilly to achieve a paste-like texture (2:1:2). Refined wheat flour (maida) was incorporated as the binding substance. The ingredients for a spice mixture included black pepper powder (0.75 per cent), cinnamon (0.05 per cent), nutmeg (0.05 per cent) and mace (0.05 per cent). The curing process involved the use of the following ingredients: sodium nitrite and sodium tripolyphosphate.

Standard culture *Pediococcus acidilactici* (MTCC 7442) was procured from the Microbial Type Culture Collection and Gene Bank (MTCC), Institute of Microbial Technology (IMTECH), Chandigarh, India and *Lactobacillus plantarum* IDK 120 (VTCDM 648 B) was procured from starter culture lab of Dairy Microbiology Division of Verghese Kurien Institute of Dairy and Food Technology (VKIDFT), Mannuthy. The casings utilised for the study were artificial-fibrous casings (Viscofan SA, Bangalore).

The control sausage batter was prepared as per the Ana et al. (2020) by grinding the meat and other components, mixing with curing ingredients, binder, starter cultures and spices until homogeneous. The batter was then stuffed into casings, pierced to release air pockets and fermented at 22°C until achieving a pH of 4.6 to 5.2. Following fermentation, the sausages were smoked for 4 hours at 20°C, then dried at 16°C until reaching a 30% weight loss, with relative humidity decreasing from 85% to 65%. Treatment consisted of addition of wheat bran at 6, 8 and 10 per cent levels each to the sausage formulation replacing the carabeef, besides other additives which were used in control at similar concentration. After drying, each product were shallow fried after slicing in meat slicer (Slicer Automatic, Model: 300 VV-CE), calculated cooking yield and kept for sensory analysis. Based on the cooking yield and sensory properties such as general appearance, flavour, texture, juiciness, sourness, aftertaste and overall acceptability on an 8-point descriptive scale where, 8 = extremely liked and 1 = extremely disliked (Keeton, 1983), the optimum levels of each dietary fibre source were identified. Then the selected treatment and control was subjected to shelf-life study.

## Analysis

The selected products (control and most acceptable treatment with dietary fibre source) were further analysed for product pH (O' Halloran et al., 1997), moisture AOAC (2016), Thiobarbituric acid reactive substance value (Witte et al., 1970), tyrosine value (Strange et al., 1977) and microbiological quality (APHA, 2015) viz., aerobic plate count (CFU/g), psychrotrophic count (CFU/g), yeast and mould count (CFU/g) and sensory evaluation. The storage studies were conducted on control and selected treatment fermented sausages. The products were packed in low-density polyethylene (LDPE) bags and stored at refrigeration temperature (4±1°C) and evaluated for 60 days at 15 days interval. All the experiments were replicated six times. The data obtained from physicochemical, microbiological and sensory characteristics of the control as well as selected treatment fermented sausages was assessed statistically by repeated measures ANOVA, oneway ANOVA and Friedmann test using the SPSS software version 24.0 (Snedecor and Cochran, 1994).

#### **Results and discussion**

The statistically analysed results for cooking yield and sensory analysis are depicted in table 1 below.

There was significant difference (p<0.01) between pH of control with treatments. Control exhibited significantly higher (p<0.01) pH. T3 exhibited significantly lower (p<0.01) pH. Significant differences (p<0.01) were observed among the cooking yields of C, T1, T2, and T3, with T3 exhibiting the highest yield and C the lowest. Sensory evaluation revealed that T2 has significantly

 Table 1. Cooking yield, product pH and sensory parameters of fermented carabeef sausage incorporated with wheat bran

Attributes	с	T1	T2	тз	F value/ ଅ² value (p-value)				
Cooking yield	66.07 <sup>d</sup> ±0.31	67.18°±0.14	68.38 <sup>b</sup> ±0.33	70.50ª±0.40	36.694 (<0.001)**				
Product pH	4.72ª±0.01	4.70 <sup>b</sup> ±0.00	4.68 <sup>b</sup> ±0.01	4.65°±0.00	19.171 (<0.001)**				
	Sensory evaluation								
Appearance and colour	7.59ª± .06	7.43 <sup>b</sup> ± 0.09	$7.64^{a} \pm 0.07$	7.43 <sup>b</sup> ± 0.09	20.483** (<0.001)				
Texture	7.47ª±0.05	7.21 <sup>b</sup> ±0.04	7.67ª±0.09	7.07 <sup>b</sup> ±0.03	48.219** (<0.001)				
Juiciness	7.29ª± 0.10	7.24ª ± 0.10	7.38ª± 0.10	7.22 <sup>b</sup> ± 0.06	20.768** (<0.001)				
Flavour	7.41ª± 0.10	7.00 <sup>b</sup> ± 0.06	7.47ª ± 0.11	6.98 <sup>b</sup> ± 0.08	37.295** (<0.001)				
Sourness	7.30 <sup>a</sup> ± 0.10	7.07 <sup>b</sup> ± 0.08	7.46ª ± 0.10	7.08 <sup>b</sup> ± 0.09	23.689** (<0.001)				
After Taste	7.36 <sup>b</sup> ± 0.09	7.16 <sup>bc</sup> ± 0.09	7.56ª± 0.13	7.01°± 0.08	38.573** (<0.001)				
Overall acceptability	7.36 <sup>b</sup> ± .09	7.17°± .08	7.74 <sup>a</sup> ± 0.09	7.07°± .08	46.500** (<0.001)				

\* Significant at 0.05 level; \*\* Significant at 0.01 level; ns – non- significant at 0.05 level. Means with different superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error. Number of observations = 24

C: Control

T<sub>1</sub>: Fermented carabeef sausage developed by incorporating 6 per cent wheat bran

T2: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

T<sub>3</sub>: Fermented carabeef sausage developed by incorporating 10 per cent wheat bran

inflated scores for aftertaste and overall acceptability. Furthermore, although there is statistical similarity with the control group regarding appearance, color, texture, flavor, juiciness, and sourness scores, T2 received a higher score and showed significant differences from other treatments. The sensory analysis suggests that T2 stands out as the best treatment, while the cooking yield results favor T3. However, since T2 exhibits a higher cooking yield than the control with best sensory scores, it is chosen as the superior treatment, with 8 per cent considered the optimal level of wheat bran inclusion.

## Dietary fibre content

There was significant difference (p<0.01) viewed in the dietary fibre values among C, T1, T2, and T3. T3 exhibited significantly highest (p<0.01) and C showed significantly (p<0.01) lowest value (Table 2).

#### Effect of storage on product pH

The dynamic changes in pH values for control and treatment fermented sausages over a 60-day period, presented in Table 3, underline the microbial activity during storage. Initially, a decrease in pH was

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noticed across all samples until day 30, featured to the fermentation of carbohydrates (specifically dextrose) by mixed bacterial cultures, resulting in lactic acid production. This is consonant with the findings of Antara *et al.* (2004), who noted that carbohydrate fermentation by lactic acid bacteria leads to a reduction in pH. Liepe and Probic (1986) also advocated that the scaling down in pH during refrigerated storage is primarily due to the activity of lactic acid bacteria.

However, after the 30th day, an escalation in pH was noted, which could be linked to the breakdown of organic acids and the formation of basic nitrogenous compounds, as explained by Kayaardi and Gok (2003) and Vural (1998). This upward trend in pH could also indicate the growth of spoilage bacteria, as remarked by Ahmad and Srivastava (2007), who reported a rise in pH by the 75<sup>th</sup> day due to such microbial activity.

#### Effect of storage on moisture content

The moisture content of control and treatment fermented sausages were assessed on days 0, 15, 30, 45 and 60 of storage and mean values expressed in percentage are displayed in table 4. On statistical analysis,

Table 2: Dietary fibre of fermented carabeef sausage incorporated with wheat bran

Parameters	С	T1	T2	Т3	F value (p value)
Dietary fibre	1.23 <sup>d</sup> ±0.01	1.85°±0.02	2.40 <sup>b</sup> ±0.04	3.16 <sup>a</sup> ±0.03	2089.632 (<0.001)**

\* Significant at 0.05 level; \*\* Significant at 0.01 level; ns – non- significant at 0.05 level. Means with different superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error. Number of observations = 24

C: Control

T,: Fermented carabeef sausage developed by incorporating 6 per cent wheat bran

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 10 per cent wheat bran

Table 3. Effect of storage period on pH of control and treatment fermented sausage

Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	$4.72^{Aa} \pm 0.01$	4.59 <sup>BC</sup> ±0.02	4.55 <sup>c</sup> ±0.03	4.62 <sup>B</sup> ±0.02	4.69 <sup>Ab</sup> ±0.02	19.402** (<0.001)
T2	4.68 <sup>Bb</sup> ±0.01	4.61 <sup>c</sup> ±0.02	4.53 <sup>D</sup> ±0.03	4.63 <sup>c</sup> ±0.02	4.74 <sup>Aa</sup> ±0.02	19.332** (<0.001)
F value (p-value)	23.085** (<0.001)	12.393** (<0.001)	3.013* (0.054)	2.868* (0.062)	4.142* (0.020)	

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

Table 4. Effect of storage period	on moisture content of	of control and treatmen	t fermented sausage
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Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	40.33 <sup>Ab</sup> ± 0.41	39.38 <sup>Bb</sup> ± 0.52	38.33 <sup>Cb</sup> ±0.49	37.18 <sup>Db</sup> ±0.70	36.28 <sup>Eb</sup> ±0.72	31.314** (<0.001)
T2	44.38 <sup>Aa</sup> ±0.41	42.75 <sup>Ba</sup> ±0.52	41.28 <sup>Ca</sup> ±0.49	40.27 <sup>Da</sup> ±0.70	39.48 <sup>Ea</sup> ±0.72	32.842** (<0.001)
F value (p-value)	22.311** (<0.001)	9.458** (<0.001)	8.539* (0.001)	4.666* (0.013)	4.320* (0.017)	

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

the moisture content of both control and selected treatment sausages decreased significantly (p<0.01) throughout the storage period. Ahmad and Srivastava (2007) also noted consistent decrease in moisture content of fermented carabeef sausages incorporated with different levels of heart and fat during refrigerated storage. The decrease in moisture content may be due to evaporation of moisture through the permeable LDPE film.

Cofrades *et al.* (2000) reported that the integration of dietary fiber into meat products promotes stronger linkage between water and fat molecules. Choi *et al.* (2008) also reported that when rice bran fiber was added to the product, the water holding capacity was higher. In their study, Ham *et al.* (2016) found that introducing collagen fiber led to increased water retention in sausages, resulting in higher yields and elevated moisture levels in the final product. So, the significantly higher moisture content of treated sausages with that of control might be due to the increased water holding capacity of fibre present in the treatment sausage.

#### Effect of storage on TBARS value

TBARS value indicate the oxidative rancidity of the product represented as mg of malonaldehyde/kg for controls and treatments throughout the storage period were presented in table 5.

Ulu (2004) reported that the TBARS value served as a measure for assessing the lipid oxidation in fermented sausages. The TBARS value of control sausage increased significantly (p<0.01) throughout the storage period. TBARS value of T2 also exhibited increasing trend and the change was not significant (p< 0.01) between 30<sup>th</sup> and 45<sup>th</sup> day. Ahmad and Srivastava (2007) also reported an increase in TBARS value of fermented carabeef sausage incorporated with different levels of heart and fat. The

Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	0.96 <sup>Ea</sup> ±0.01	1.10 <sup>D</sup> ±0.03	1.38 <sup>cb</sup> ±0.11	1.90 <sup>B</sup> ±0.11	2.20 <sup>Ab</sup> ±0.04	205.674** (<0.001)
T2	0.77 <sup>Db</sup> ±0.01	1.07 <sup>c</sup> ±0.03	1.50 <sup>Ba</sup> ±0.11	1.63 <sup>B</sup> ±0.11	2.37 <sup>Aa</sup> ±0.04	334.245** (<0.001)
F value (p-value)	68.646** (<0.001)	5.218* (0.008)	6.092* (0.004)	15.526** (<0.001)	126.272** (<0.001)	

Table 5. Effect of storage period on TBARS value of control and treatment fermented sausage

\*\*Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

Table 6. Effect of storage period on Tyrosine value of control and treatment fermented sausages

Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	37.93 <sup>⊑</sup> ±4.51	64.41 <sup>Da</sup> ±2.23	74.46 <sup>Ca</sup> ±1.90	84.53 <sup>Ba</sup> ±2.42	83.36 <sup>Aa</sup> ±0.57	72.682** (<0.001)
T2	31.16 <sup>E</sup> ±4.51	50.77 <sup>Db</sup> ±2.23	66.17 <sup>Cb</sup> ±1.90	73.83 <sup>Bb</sup> ±2.42	83.82 <sup>Ab</sup> ±0.57	118.384** (<0.001)
F value (p-value)	1.439 <sup>ns</sup> (0.261)	10.020* (0.002)	17.822** (<0.001)	10.438** (<0.001)	51.927** (<0.001)	

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

Table 7. Effect of storage period on TVC of control and treatment fermented sausages

Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	5.67 <sup>D</sup> ±0.18	5.68 <sup>D</sup> ±0.10	6.34 <sup>c</sup> ±0.12	7.39 <sup>B</sup> ±0.12	8.45 <sup>A</sup> ±0.09	83.004** (<0.001)
T2	5.42 <sup>D</sup> ±0.18	5.76 <sup>D</sup> ±0.10	6.67 <sup>c</sup> ±0.12	7.40 <sup>B</sup> ±0.12	8.74 <sup>A</sup> ±0.09	93.233** (<0.001)
F value (p-value)	1.181 <sup>ns</sup> (0.342)	0.143 <sup>ns</sup> (0.933)	1.503 <sup>ns</sup> (0.244)	0.254 <sup>ns</sup> (0.857)	2.061 <sup>ns</sup> (0.138)	

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

decrease in TBARS value of T2 may be associated with the antioxidant properties of fibre source involved. However, the TBARS value of both control and treatment sausages in the current study fell within the acceptable level. The acceptable TBARS range for fermented sausages is 0.6 to 2.8 mg MDA/kg (Marco *et al.*, 2006).

# Effect of storage on Tyrosine Value

The analysis of TV, which serve as an indicator of proteolysis within the product, expressed as mg of tyrosine per 100g of sausage, was tabulated for control and treatment sausages (T2) over a storage period of 60 days. The results, as depicted in Table 6, reveal a significant increase (p<0.01) in the TV for both control and treatment sausages throughout the storage period. This finding aligns with Nie *et al.* (2014), who, through SDS-PAGE analysis, manifested that lactic acid bacteria (LAB) could promote the breakdown of myofibrillar and sarcoplasmic proteins, leading to increased levels of tyrosine.

On the initial day of storage, no significant difference (p>0.05) was found in the TVs among C and T2, indicating a uniform start with regards to proteolytic activity. However, as storage progressed, the TV of the control differed significantly from those of the treatment sausage (p<0.05). The observed variance in TVs between control and treatment sausages could be remarked for the presence of fiber sources in the treatments, which may enhance the growth of LAB and thereby increase proteolytic activity.

Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	2.67 <sup>c</sup> ±0.08	3.25 <sup>B</sup> ±0.06	3.17 <sup>Ba</sup> ±0.06	4.08 <sup>Ab</sup> ±0.04	4.09 <sup>A</sup> ±0.04	137.995** (<0.001)
T2	2.67 <sup>D</sup> ±0.08	3.29 <sup>c</sup> ±0.06	3.34 <sup>Cb</sup> ±0.06	4.43 <sup>Aa</sup> ±0.04	4.13 <sup>в</sup> ±0.04	189.025** (<0.001)
F value (p-value)	0.486 <sup>ns</sup> (0.696)	0.392 <sup>ns</sup> (0.760)	10.911** (<0.001)	14.639** (<0.001)	9.876** (<0.001)	

Table 8. Effect of storage period on psychrotrophic count of control and treatment fermented sausages

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

Table 9.	Effect of sto	rage period or	yeast and	mould count of	of control a	nd treatment	fermented	sausages
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Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	F value (p-value)
С	1.40 <sup>Eb</sup> ± 0.29	2.70 <sup>Db</sup> ±0.12	2.98 <sup>cb</sup> ±0.16	3.41 <sup>вь</sup> ±0.13	3.77 <sup>Ab</sup> ±0.10	55.059** (<0.001)
T2	2.40 <sup>Ea</sup> ±0.29	3.21 <sup>Da</sup> ±0.12	3.60 <sup>Ca</sup> ±0.16	3.93 <sup>Bba</sup> ±0.13	4.28 <sup>Aa</sup> ±0.10	54.570** (<0.001)
F value (p-value)	3.252* (0.043)	8.115* (0.001)	3.757* (0.27)	5.282* (0.008)	15.350** (0.001)	

\*\* Significant at 0.01 level. \* Significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

## Effect of storage on microbiological quality

Statistical analysis denoted that the TVC of both control and treatment fermented sausages experienced an increasing trend throughout the storage period. However, there was no significant difference (p > 0.05) between TVC of control and treatment sausages throughout the storage period (Table 7).

Similar studies on fermented carabeef sausages was conducted by Ahmad and Srivastava (2007). They analysed that during refrigerated storage, a significant (p < 0.05) linear augment in the TVC was observed for control and treatments as storage progressed beyond 30 days. They mentioned that when total viable counts on meat tissue exceeds 7 log<sub>10</sub>/g, off odour and slime starts. At the 60<sup>th</sup> day of storage we got 7 log cfu/g colonies, this count includes spoilage bacteria along with inoculated LAB. And no indication for off odour and slime growth could be observed at the same day. Supportively Smith and Palumbo, (1983) reported that LAB inhibit other pathogenic and spoilage microorganism hence we assume that eventhough the total viable count exceeded the limit it might be due to the Lactic acid bacterial count.

Statistical analysis showed that the psychrotrophic count of both control and T2 increased throughout the storage (Table 8). On  $60^{th}$  day of storage count were in the range of 4.09 and 4.13 for C, and T2 respectively. Our result was not in accordance of Qureshi *et al.*, (2022) they suggested that psychrotrophic bacterial count should not exceed  $10^3$  CFU/g. Our elevated psychrotrophic count might be due to the addition of stater culture inoculum.

The analysis of yeast and mould counts in control and treatment fermented sausages, indicated a significant increase (p<0.01) in these counts over the storage period (Table 9). By the 60th day, the counts had reached levels of 3.77 and 4.28 for C and T2 respectively. According to Qureshi *et al.* (2022), yeast and mould counts exceeding  $10^4$  CFU/g indicate spoilage, situating the observed counts within a range indicative of spoilage. Paramithiotis and Drosinos (2010) added that while spices and additives generally keep the yeast-mould population below the detection limit, in some instances, these populations can become dominant, citing instances where whole pepper and paprika harbored yeast and mould populations of 6.72 and 5.8 log<sub>10</sub> CFU/g, respectively.

Sampla			Storage days		Storage days						
Sample	0 <sup>th</sup> day	15 <sup>th</sup> day	30 <sup>th</sup> day	45 <sup>th</sup> day	60 <sup>th</sup> day	Overall mean					
		Арр	earance and co	lour							
С	7.32 <sup>Ab</sup> ±0.08	7.50 <sup>A</sup> ±0.08	7.39 <sup>Aa</sup> ±0.08	7.05 <sup>Ba</sup> ±0.03	6.75 <sup>Ca</sup> ±0.08	7.20±0.04					
T2	7.55 <sup>Aa</sup> ±0.07	7.42 <sup>B</sup> ±0.03	7.04 <sup>Cb</sup> ±0.02	6.32 <sup>Db</sup> ±0.04	$6.35^{\text{Db}} \pm 0.03$	6.93±0.05					
Overall mean	7.51±0.04	7.47±0.03	7.23±0.04	6.84±0.05	6.59±0.04						
			Texture								
С	$7.20^{Ab} \pm 0.08$	7.20 <sup>Ab</sup> ±0.08	7.19 <sup>Aa</sup> ±0.08	6.54 <sup>Ba</sup> ±0.10	$6.49^{Ba} \pm 0.09$	6.92±0.05					
T2	7.76 <sup>Aa</sup> ±0.08	7.76 <sup>Aa</sup> ±0.08	7.00 <sup>Bb</sup> ±0.00	6.32 <sup>Cb</sup> ±0.04	$5.96^{\text{Db}} \pm 0.04$	6.96±0.08					
Overall mean	7.51±0.04	7.36±0.04	7.05±0.03	6.4±0.03	6.21±0.04						
			Juiciness								
С	$7.25^{Ab} \pm 0.09$	6.96 <sup>Bb</sup> ±0.07	6.98 <sup> в</sup> ±0.13	6.74 <sup>Ca</sup> ±0.09	6.29 <sup>Ca</sup> ±0.05	6.84±0.05					
T2	7.49 <sup>Aa</sup> ±0.09	7.45 <sup>Aa</sup> ±0.06	7.11 <sup> в</sup> ±0.03	6.04 <sup>Cb</sup> ±0.10	5.93 <sup>Cb</sup> ±0.03	6.80±0.07					
Overall mean	7.43±0.04	7.11±0.05	7.04±0.05	6.24±0.05	6.11±0.03						
			Flavour								
С	7.56 <sup>A</sup> ±0.08	7.17 <sup>Bb</sup> ±0.03	7.10 <sup> в</sup> ±0.16	6.52 <sup>Ca</sup> ±0.09	6.23 <sup>Da</sup> ±0.06	6.91±0.06					
T2	7.60 <sup>A</sup> ±0.07	7.52 <sup>Aa</sup> ±0.05	7.05 <sup> в</sup> ±0.02	6.02 <sup>Cb</sup> ±0.09	6.01 <sup>Cb</sup> ±0.07	6.84±0.07					
Overall mean	7.57±0.03	7.30±0.03	6.94±0.06	6.21±0.04	6.09±0.03						
			Sourness								
С	7.50 <sup>A</sup> ±0.07	7.18 <sup>Bb</sup> ±0.04	6.94 <sup>Bb</sup> ±0.15	6.50 <sup>Ca</sup> ±0.09	6.54 <sup>Ca</sup> ±0.15	6.93±0.06					
T2	7.61 <sup> ^</sup> ±0.05	7.35 <sup>Ba</sup> ±0.07	7.31 <sup>Ba</sup> ±0.03	5.99 <sup>Cb</sup> ±0.10	$6.10^{Cb} \pm 0.07$	6.87±0.07					
Overall mean	7.57±0.03	7.26±0.03	7.13±0.06	6.21±0.05	6.15±0.05						
			After taste								
С	7.18 <sup>Ab</sup> ±0.08	7.00 <sup>Ab</sup> ±0.07	7.01 <sup>Ab</sup> ±0.12	6.43 <sup>Ba</sup> ±0.09	6.27 <sup>Ba</sup> ±0.09	6.78±0.05					
T2	7.76 <sup>Aa</sup> ±0.08	$7.52^{Bab} \pm 0.05$	7.35 <sup>Ca</sup> ±0.04	6.11 <sup>Db</sup> ±0.07	$5.88^{Eb} \pm 0.08$	6.92±0.08					
Overall mean	7.49±0.05	7.23±0.04	7.02±0.05	6.24±0.04	6.00±0.05						
		Ov	verall acceptabi	lity							
С	$7.37^{Ab} \pm 0.05$	7.13 <sup>Bb</sup> ±0.04	7.12 <sup>Bb</sup> ±0.10	6.57 <sup>Ca</sup> ±0.07	6.20 <sup>Da</sup> ±0.05	6.88±0.05					
T2	7.77 <sup>Aa</sup> ±0.07	7.37 <sup>Ba</sup> ±0.06	7.37 <sup>Ba</sup> ±0.03	6.37 <sup>Cb</sup> ±0.03	$5.85^{\text{Db}} \pm 0.06$	6.95±0.07					
Overall mean	7.60±0.03	7.25±0.03	7.17±0.03	6.38±0.03	6.00±0.03						

Table 10. Effect of storage period on sensory attributes of control and treatment fermented sausages

\*\* Significant at 0.01 level. \* Significant at 0.05 level. ns: non-significant at 0.05 level. Means with different uppercase superscripts in rows have significant difference. The values are expressed as their Mean ± Standard error.

C: Control fermented carabeef sausage

T<sub>2</sub>: Fermented carabeef sausage developed by incorporating 8 per cent wheat bran

#### Effect of storage on sensory evaluation

Sensory attributes particularly for appearance and colour, texture, juiciness, flavour, sourness, aftertaste and overall acceptability for C and T2 progressively declined with the progress of storage period (Table 10).

By analysing the statistical values of TBARS value and TV, we could assume that it has an inverse correlation with sensory score values. These declining sensory values might be due to lipid rancidity or proteolysis of product thus affecting the sensory values adversely. Our study was in accordance with Ahmad and Srivastava (2007), in their study also quality deterioration of product was observed across the storage period and product shelf life was also 60 days at refrigeration temperature  $(4\pm 1^{\circ}C)$ .

## Conclusion

In this study, we aimed to develop and assess wheat bran-enriched fermented carabeef sausages, focusing on their physico-chemical, sensory and microbiological characteristics, as well as their refrigerated storage stability at  $4\pm1$ °C. Three sausage variants (T1, T2 and T3) were prepared by substituting 6%, 8% and 10% of the carabeef with wheat bran, respectively. The T2 variant, containing 8% wheat bran, was identified as optimal based on cooking yield and sensory evaluations. All treatment sausages showed improved cooking yields and a higher fibre content compared to the control, with notable differences in moisture content and pH levels during storage. Over a 60-day period, both control and T2 sausages demonstrated a decrease in moisture content and a temporary reduction in pH, followed by a subsequent increase. The oxidative and nitrogenous degradation. as indicated by TBARS and Tyrosine values, escalated significantly in the control sausages. Similarly, microbial counts, including total viable count, psychrotrophs, yeast, and mould, showed a significant increase in both sausage types over time. Despite the sensory attributes declining over the storage period, the T2 sausages maintained acceptable levels of quality up to 60 days. This study supports the potential of wheat bran as a viable fibre source in enhancing the nutritional profile of fermented sausages without compromising their overall sensory acceptability and storage stability.

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# **Conflict of interest**

The authors declare that they have no conflict of interest.

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